Identification of deoxyribonuclease II as an endonuclease involved in apoptosis.


Source

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Abstract

Cell death occurs by apoptosis during programmed deletion of cells and following exposure to cytotoxic agents. Central to the mechanism of apoptosis is internucleosomal DNA digestion by an endogenous endonuclease which is thought to mediate cell death. An axiom of apoptosis is that the endonuclease involved is a Ca2+/Mg(2+)-dependent endonuclease. During purification of endonucleases from Chinese hamster ovary cells, we found little Ca2+/Mg(2+)-dependent endonuclease activity, but large amounts of an endonuclease active below pH 7. This acidic endonuclease was activated in intact cells by reducing intracellular pH values below 7 with a proton ionophore. This activity generated internucleosomal digestion of DNA characteristic of apoptosis. Nuclear extracts contained a cation-independent endonuclease with identical pH-dependent activity. We have compared the acidic endonuclease to bovine deoxyribonuclease II (DNase II) and have found them nearly identical by all tests, including sensitivity to various inhibitors, purification by the same chromatographic steps, and recognition by antibody raised against the bovine enzyme. Addition of either the acidic endonuclease or bovine DNase II to isolated nuclei induced internucleosomal DNA digestion up through pH 6.5. These data demonstrate that DNase II can mediate internucleosomal DNA digestion characteristic of apoptosis following intracellular acidification. Furthermore, these data question the premise that the Ca2+/Mg(2+)-dependent endonuclease is the only endonuclease involved in apoptosis.

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