Apoptotic insults to human chondrocytes induced by sodium nitroprusside are involved in sequential events, including cytoskeletal remodeling, phosphorylation of mitogen-activated protein kinase kinase kinase-1/c-Jun N-terminal kinase, and Bax-mitochondria

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摘要

Abstract

Nitric oxide (NO) can regulate chondrocyte activities. This study was aimed to evaluate the molecular mechanisms of NO donor sodium nitroprusside (SNP)-induced insults to human chondrocytes. Exposure of human chondrocytes to SNP increased cellular NO levels but decreased cell viability in concentration- and time-dependent manners. SNP time dependently induced DNA fragmentation and cell apoptosis. Treatment with 2-phenyl-4,4,5,5-tetramethyl-imidazoline-1-oxyl 3-oxide, an NO scavenger, significantly lowered SNP-induced cell injuries. Administration of SNP interrupted F-actin and microtubule cytoskeletons and stimulated phosphorylation of mitogen-activated protein kinase kinase kinase-1 (MEKK1) and c-Jun N-terminal kinase (JNK). Similar to SNP, cytochalasin D, an inhibitor of F-actin formation, disturbed F-actin polymerization and increased MEKK1 and JNK activations. Overexpression of a dominant negative mutant of MEKK1 (dnMEK1) in human chondrocytes significantly ameliorated SNP-induced cell apoptosis. Exposure to SNP promoted Bax translocation from the cytoplasm to mitochondria, but application of dnMEKK1 lowered the translocation. SNP time dependently decreased the mitochondrial membrane potential, complex I NADH dehydrogenase activity, and cellular ATP levels, but increased the release of cytochrome c from mitochondria to the cytoplasm. Activities of caspase-9, -3, and -6 were sequentially increased by SNP administration. This study shows that SNP can induce apoptosis of human chondrocytes through sequential events, including cytoskeletal remodeling, activation of MEKK1/JNK, Bax translocation, mitochondrial dysfunction, cytochrome c release, caspase activation, and DNA fragmentation.