

# **Methylglyoxal-induced fibronectin gene expression through Ras-mediated NADPH oxidase activation in renal mesangial cells.**

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## **Abstract**

**BACKGROUND:** The formation of methylglyoxal (MGO), a highly reactive dicarbonyl compound, is accelerated under diabetic conditions. Although recent studies have suggested that apoptotic cell death is involved in diabetic nephropathy, the precise mechanism of MGO-induced renal fibrosis remains to be elucidated. **METHODS:** Rat kidney mesangial cells with or without pretreatment with inhibitors, including superoxide dismutase, catalase, L-NAME, diphenylene iodonium, rotenone, allopurinol, PD98059, SB203580 and SP600125 were cultured in medium containing 100 microM MGO. In the MGO-treated cell culture system, fibrosis-related signalling pathway was assessed by enzyme-linked immunosorbent assay, reverse transcription-polymerase chain reaction and western blotting. **RESULTS:** Expression of fibronectin induced by MGO was highest after 48 h treatment. Superoxide production rapidly increased after 2 h and remained at a high level for 24 h. Scavenging O(2) (-) reversed transforming growth factor beta 1 (TGF-beta1) and fibronectin mRNA level. Pretreatment with diphenylene iodonium significantly suppressed MGO-induced superoxide, TGF-beta1 expression and fibronectin gene expression, indicating that NADPH oxidase is responsible for inducing superoxide formation and subsequently induced renal fibrosis. High MGO rapidly enhanced Ras activation in 1 h and progressively increased cytosolic p38 activation. Additionally, SB203580 pretreatment reduced MGO promotion of fibronectin gene activation suggesting that cytosolic p38 activation might affect MGO-induced renal mesangial fibrosis. Inhibiting Ras activity with manumycin A significantly reduced the promoting effect of MGO on superoxide synthesis, and fibronectin expression. **CONCLUSION:** Induction of superoxide by Ras via p38 pathway activates fibrotic gene transcription of mesangial cells. Reduction of oxidative stress by scavenging superoxide may offer an alternative strategy for controlling MGO-induced renal fibrosis.