Oxido nítrico e bezafibrato.

O bezafibrato aumenta a expressão gênica do oxido nítrico sintase e aumenta o oxido nítrico endotelial: benefícios na hipertensão arterial, na aterosclerose além do efeito hipolipemiante. Cuidado: o bezafibrato provoca muitos efeitos colaterais, parte deles devido à inibição da geração mitocondrial de ATP.

José de Felipe Junior

Bezafibrate up-regulate endothelial nitric oxide gene expressions via peroxisome proliferator-activated receptors alpha-dependent and independent pathways in cultured Bovine endothelial cells


Cardiovascular Division, Internal Medicine Department, Institute of Hypertension, Tongji Hospital, Huazhong University of Science and Technology, Wuhan 430030, China.

Abstract

OBJECTIVE: To investigate the effects and related mechanisms of bezafibrate, a ligand of peroxisome proliferator-activated receptors alpha (PPARalpha), on endothelial nitric oxide synthase (eNOS). METHODS: Firstly, in cultured bovine aorta endothelial cells (BAEC), the effects of bezafibrate on eNOS mRNA and protein levels were investigated by Northern blot and Western blot. The half-life of eNOS mRNA and NO production determined from BAECs treated with bezafibrate were performed by real-time quantitative PCR and NO assay. Next, the effects of bezafibrate on signal pathways in BAEC, through inhibitors of PPARalpha, PI3 kinase and MAPK, were investigated by Western blot. Then luciferase reporter gene driven by human eNOS promoter were cloned and transfected endothelial cells to see the effects of bezafibrate on eNOS promoter-driven luciferase activity. RESULTS: In cultured BAEC, bezafibrate significantly upregulated eNOS expressions at protein and mRNA levels in a concentration-dependent fashion (50 - 200 micromol/L) (P < 0.05), and increased nitric oxide (NO) production, respectively (control (14.97 +/- 1.29) micromol/L, different concentration groups (25.12 +/- 1.25) micromol/L, (30.12 +/- 1.85) micromol/L, (33.47 +/- 1.22) micromol/L), and enhanced phosphorylation of eNOS-ser-1179 site (P < 0.05), but not eNOS-thr-497 site. Through real-time quantitative PCR, bezafibrate prolonged eNOS mRNA half-life from 3.1 hour to 6.1 hour were demonstrated. Further studies showed that bezafibrate treatments significantly enhanced dose-dependently luciferase activity (relative luciferase activity in 100 micromol/L and 200 micromol/L groups 4429.43 +/- 391.41 and 6187.5 +/- 307.53), compared with untreated luciferase reporter gene group (3361.81 +/- 316.85) (P < 0.05 and P < 0.01, respectively), and induced MAPK phosphorylation expression (P < 0.05 and P < 0.01, respectively). Then these effects of bezafibrate upregulated eNOS expressions could be blocked by PPARalpha antagonist, MAPK and PI3K inhibitor while not affected by PKC and MEK inhibitor (P < 0.01). CONCLUSIONS: Bezafibrate can upregulate eNOS expression, enhance eNOS-ser-1179 site phosphorylation, increase NO production and transcription level and stability of eNOS mRNA through pathway dependent of PPARalpha and nongenomic effects of bezafibrate on eNOS mRNA and NO production determined from BAEC treated with bezafibrate were performed by real-time quantitative PCR and NO assay. CONCLUSIONS: Bezafibrate can upregulate eNOS expression,

Effects of bezafibrate on the expression of endothelial nitric oxide synthase gene and its mechanisms in cultured bovine endothelial cells.


Cardiovascular Division of Internal Medicine, Department and The Institute of Hypertension, Tongji Hospital, Tongji Medical College of Huazhong University of Science & Technology, Wuhan 430030, People's Republic of China.

Abstract

OBJECTIVE: Peroxisome proliferator-activated receptors alpha (PPARalpha) is a target gene for atherosclerosis and cardiovascular diseases. However, effects of PPARalpha on endothelial nitric oxide synthase (eNOS) remain unknown. We investigated the eNOS regulation by bezafibrate, a ligand of PPARalpha, and involved signaling pathways. METHODS AND RESULTS: Firstly, in cultured bovine aorta endothelial cells (BAEC), bezafibrate significantly upregulated eNOS at protein, mRNA levels and NO production, respectively, in a concentration-dependent fashion (50-200mM). Next, the effects of bezafibrate on signal pathways and eNOS mRNA stability in BAEC were investigated. Results showed that bezafibrate induced phosphorylation of MAPK. Inhibitors of PPARalpha, PI3 kinase and MAPK, respectively, markedly attenuate bezafibrate-induced upregulation of eNOS. Bezafibrate incubation increased eNOS mRNA half-life, activated eNOS promoter, enhanced phosphorylation of eNOS ser-1179 site, and decreased phosphorylation of eNOS thr-497 site via activating ERK and Akt. CONCLUSIONS: Bezafibrate can upregulate eNOS expression, enhance phosphorylation of eNOS ser-1179, increase NO production and transcription level and stability of eNOS mRNA through pathway dependent of PPARalpha and nongenomic effects mediated by MAPK and PI3K pathways. Hence, PPARalpha ligands exert direct benefits on vessel endothelial functions through an increase in eNOS expression level and phosphorylation of eNOS ser-1179. This mechanism provides additional anti-atherosclerotic and anti-hypertension benefits of bezafibrate in addition of lipid-lowering effects.

PMID: 16256120