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SIRT1/UCP2通路在白藜芦醇抑制血管内皮细胞氧化应激损伤中的作用

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Title: Role of SIRT1/UCP2 signaling pathway in resveratrol-induced inhibition of oxidative injury in vascular endothelial cells

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关键词: 动脉粥样硬化; 白藜芦醇; 人脐静脉内皮细胞; 氧化应激; SIRT1; UCP2

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摘要: 目的 观察白藜芦醇对血管内皮细胞氧化应激损伤的抑制作用,并围绕SIRT1/UCP2信号通路探讨其作用机制。 方法 原代培养人脐静脉内皮细胞(human umbilical vein endothelial cells,HUVECs),用叔丁基过氧化氢(t-BHP)建立HUVECs氧化应激损伤模型。CCK-8法检测细胞增殖,确定半抑制浓度(IC₅₀)。荧光分光光度计检测细胞内活性氧(ROS)生成。CCK-8法检测白藜芦醇对t-BHP诱导的HUVECs活力的影响;qRT-PCR法检测白藜芦醇对SIRT1和UCP2 mRNA表达的影响;Western blot法检测白藜芦醇对SIRT1、UCP2和Caspase-3在细胞内蛋白表达的影响。 结果 t-BHP明显抑制细胞活力,IC₅₀为80 μmol/L。白藜芦醇(0.1、1、10 μmol/L)预处理2 h能显著抑制t-BHP诱导的细胞活力下降(P<0.05),并抑制t-BHP诱导的细胞内ROS增加(P<0.05)。同时,白藜芦醇能明显抑制t-BHP诱导的SIRT1的mRNA和蛋白表达下降、UCP2的mRNA和蛋白表达升高及Caspase-3表达增加(P<0.05),而Sirt1抑制剂尼克酰

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胺能削弱白藜芦醇的抑制作用。 结论 白藜芦醇可能通过活化SIRT1抑制UCP2表达, 削弱t-BHP诱导的细胞内ROS生成, 从而抑制血管内皮细胞氧化应激损伤。

Abstract: **Objective** To assess the role of silent mating type information regulation 2 homolog 1/uncoupling protein 2 (SIRT1/UCP2) signaling pathway in the inhibition of oxidative damage in vascular endothelial cells by resveratrol. **Methods** Human umbilical vein endothelial cells (HUVECs) were isolated and primarily cultured. The obtained cells were treated by tert-butyl hydroperoxide (t-BHP) at the concentrations of 20, 30, 40, 50, 60, 70, 80, 90 and 100 $\mu\text{mol/L}$ respectively for 24 h, and CCK-8 assay was used for cell viability to determine the IC_{50} value. The intracellular level of reactive oxygen species (ROS) was observed by fluorospectrophotometry. The expression of SIRT1 and UCP2 at mRNA level was determined by qRT-PCR assay. The protein expression of SIRT1, UCP2 and Caspase-3 were determined by Western blot assay. **Results** Cell viability was significantly inhibited by t-BHP treatment ($P<0.05$) and the IC_{50} value was identified as 80 $\mu\text{mol/L}$. Pretreatment with resveratrol at the doses of 0.1, 1, and 10 $\mu\text{mol/L}$ significantly inhibited the decrease of cell viability ($P<0.05$) and the increase of intracellular ROS level induced by t-BHP. Furthermore, the mRNA and protein expression of SIRT1 were up-regulated by resveratrol pretreatment, and that of UCP2 was down-regulated, yet which was abolished by niacinamide pretreatment. **Conclusion** Resveratrol may suppress the expression of UCP2 by activating SIRT1, and attenuate the increase of intracellular ROS level induced by t-BHP, and thus, inhibit t-BHP-induced endothelial injury.

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