The role of uric acid in the formation of atherosclerotic foam cells and its mechanisms

Abstract

[Objective]

1. To observe the effect of soluble uric acid and sodium urate on macrophage RAW264.7 and to determine the form of action of uric acid affecting macrophage RAW264.7.

2. To investigate the effect of sodium urate on macrophage RAW264.7 lipid accumulation and inflammation and its mechanism.

[Methods]

1. The optimal concentration of oxidized low-density lipoprotein (ox-LDL) on macrophages RAW264.7 using the Oil Red "O" staining kit and real-time quantitative fluorescence PCR (qPCR) assay, and the optimal time of action of sodium urate on macrophages RAW264.7 using the Oil Red "O" staining kit The optimal concentration and duration of action of uric acid on macrophage RAW264.7 were determined using the Oil Red "O" staining kit and qPCR experiments to determine the optimal concentration and duration of action of sodium urate on macrophage RAW264.7: Control group, 20 mg/L ox-LDL group, 15 mg/dL sodium urate group and 15 mg/dL sodium urate and 20 mg/L ox-LDL combination group. control group was incubated with high sugar DMEM medium for 72 h, 20 mg/L ox-LDL group was incubated with high sugar DMEM medium for 48 h and then given 20 mg/L The group was given high glucose DMEM medium containing 20 mg/L ox-LDL for 24 h, the 15 mg/dL sodium urate group was given high glucose DMEM medium containing 15 mg/dL sodium urate for 72 h, and the combined 15 mg/dL sodium urate and 20 mg/L ox-LDL group was given high glucose DMEM medium containing 15 mg/dL sodium urate for 48 h and then 15 mg/dL sodium urate and 20 mg/L ox-LDL. The group was given high glucose DMEM medium containing 15 mg/dL sodium urate and 20 mg/L ox-LDL for 24 h.

2. Macrophage RAW264.7 lipid accumulation and its mechanism of action using Oil Red "O" staining kit, flow cytometry and qPCR assays.

3. Exploration of macrophage RAW264.7 inflammatory factor expression levels using ELISA kits and qPCR assays.

4. Transcriptome sequencing to find differentially expressed genes.

[Results]

1. Both 15 mg/L and 20 mg/L ox-LDL induced lipid accumulation in macrophages RAW264.7 for 24 h (P<0.01), while the effect was more significant in the 20 mg/L ox-LDL group (P<0.001). 15 mg/L and 20 mg/L ox-LDL induced a decrease in SR B1 mRNA expression was decreased in both 15 mg/L and 20 mg/L ox-LDL in macrophages RAW264.7 for 24 h (P<0.001), while 20 mg/L ox-LDL in macrophages RAW264.7 for 24 h also caused an increase in CD36 mRNA expression (P<0.05).

2. Instead of increasing macrophage lipid accumulation, 30 mg/dL uric acid applied to macrophage RAW264.7 for 24 h and 36 h decreased macrophage lipid accumulation (P<0.05); 15 mg/dL sodium urate applied to macrophage RAW264.7 for 24 h and 72 h caused an increase in CD36 and TNF-α mRNA expression was increased by 15 mg/dL sodium urate in macrophages RAW264.7 for 24 h and 72 h (P<0.01 and P<0.001, respectively), and in addition, 15 mg/dL sodium urate in macrophages RAW264.7 for 72 h decreased the mRNA expression level of SR-B1 (P<0.01).

3. Significantly increased lipid accumulation in macrophages (P<0.001), mRNA expression of TNF-α in macrophages (P<0.01) and TNF-α concentration in cell culture supernatants (P<0.01) in the combined 15 mg/dL sodium urate and 20 mg/L ox-LDL group compared to the 20 mg/L ox-LDL group.

4. There were 17 differential genes expressed between the groups, of which three were non-coding genes. qPCR experiments on 14 coding genes verified that mRNA expression of the Plin2 gene and Esd gene in macrophages was significantly increased (P<0.001) in the 20 mg/L ox-LDL group compared to the Ctrl group and in the combined effect of 15 mg/dL sodium urate and 20 mg/L ox-LDL, with the increase in the combined effect of 20 mg/L ox-LDL group The increase was even more significant in the 20 mg/L ox-LDL combination group.

[Conclusion]

Soluble uric acid had no significant effect on macrophage RAW264.7 lipid accumulation, whereas sodium urate promoted macrophage RAW264.7 lipid accumulation and inflammatory factor release through upregulation of Plin2 and Esd gene expression.

[Key words]

atherosclerosis; lipids; inflammation; monosodium urate; RAW264.7 cells