Effect of Circhipk3 on Polarization of Microglial Cells in Nerve Injury Caused by Heat Radiation

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Research

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Abstract

Background: Circular RNAs play an important regulatory role in the occurrence and development of nerve damage and related diseases, but there is little research on non-coding RNAs in heat sickness, especially circRNAs, and there has been no report on the mechanism of fever progression.

Methods: 1. Mice were randomly divided into a control group, a heat radiation disease 0.8 h group (HS 0.8), 8 h group (HS 8), 24 h group (HS 24). By establishing a mouse model of heat shock (HS), heat-damaged brain tissue was obtained, microglia were isolated. 2. QPCR was used to detect M1 and M2 marker molecules in microglia, and to evaluate the polarization direction and type of microglia. 3. The expression level of circhipk3 in microglial cells, and the effect of circhipk3 on microglial polarization was determined by determining the expression of circhipk3 in microglial cells.

Results: The expressions of CD45 and CD11-b in the HS 8 group were significantly higher than those in the normal group, while the expressions of CD45 and CD11-b in the HS 24 group were significantly lower than those in the HS 8 group. At the same time, the expression of CD206, FIZZ, and Arg1 in the HS 8 group began to increase compared with the normal group, while CD206, FIZZ, and Arg1 in the HS 24 group significantly increased when compared with the normal group. Circhipk3 mimics significantly increased the expression of Arg1 and inhibited the expressions of CD45 and HO-1, while the circhipk3 inhibitor promoted the expressions of CD45 and HO-1 and inhibited the expression of Arg1.

Conclusion: Microglial cells were the main M1-type in early neurological injury of heat radiation disease. HO-1 may be one of the microglial M1-type markers. The high expression of circhipk3 in microglial cells mainly promoted its transformation to the M2 type.

Full Text

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Figures

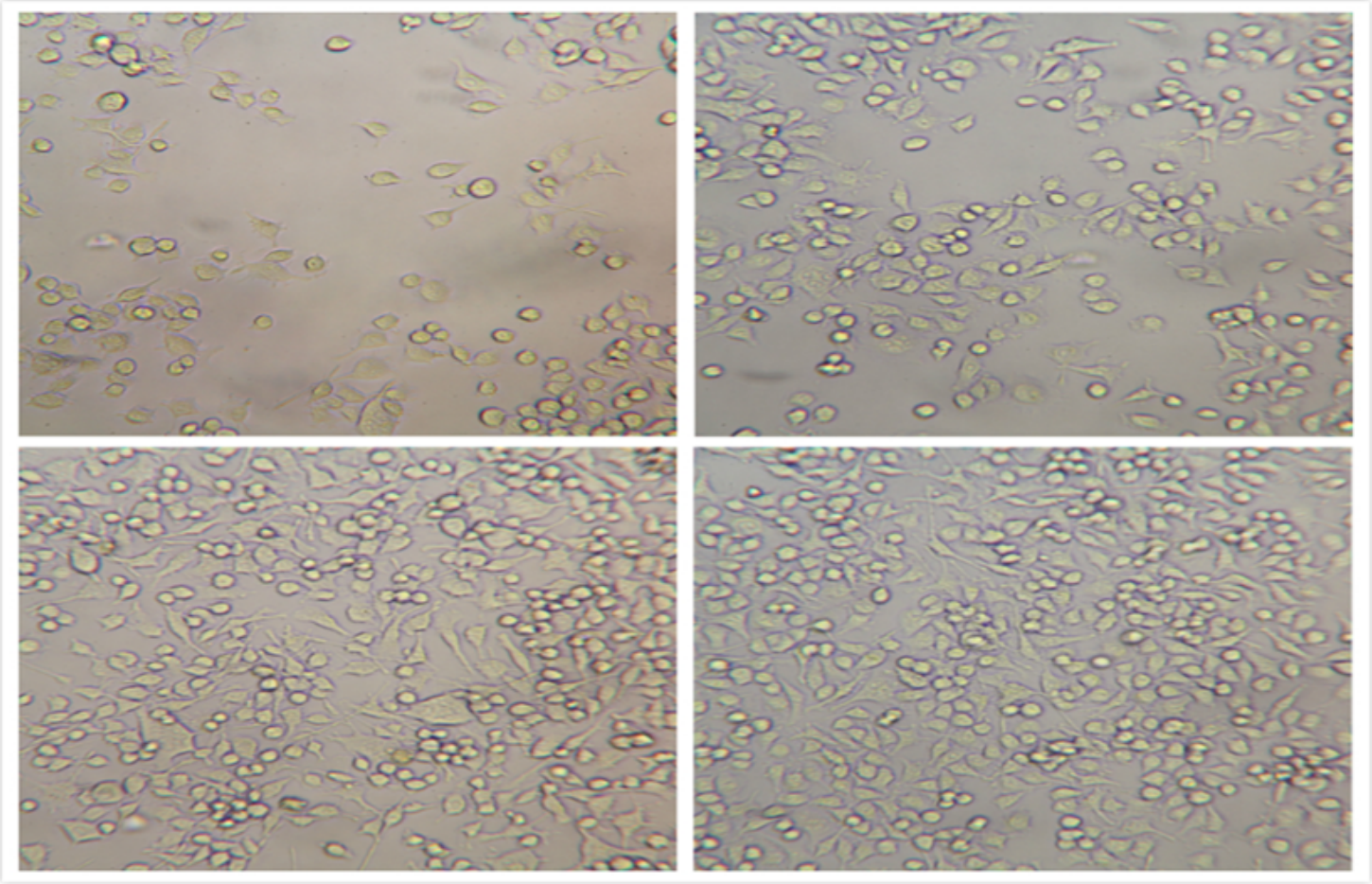

Figure 1

Microglial changes in mice after thermal stimulation. A: Control group; B: Heat stroke (HS) 0.8 group; C: HS 8 group; D: HS 24 group. Electron micrographs of mouse microglia at different times after thermal stimulation

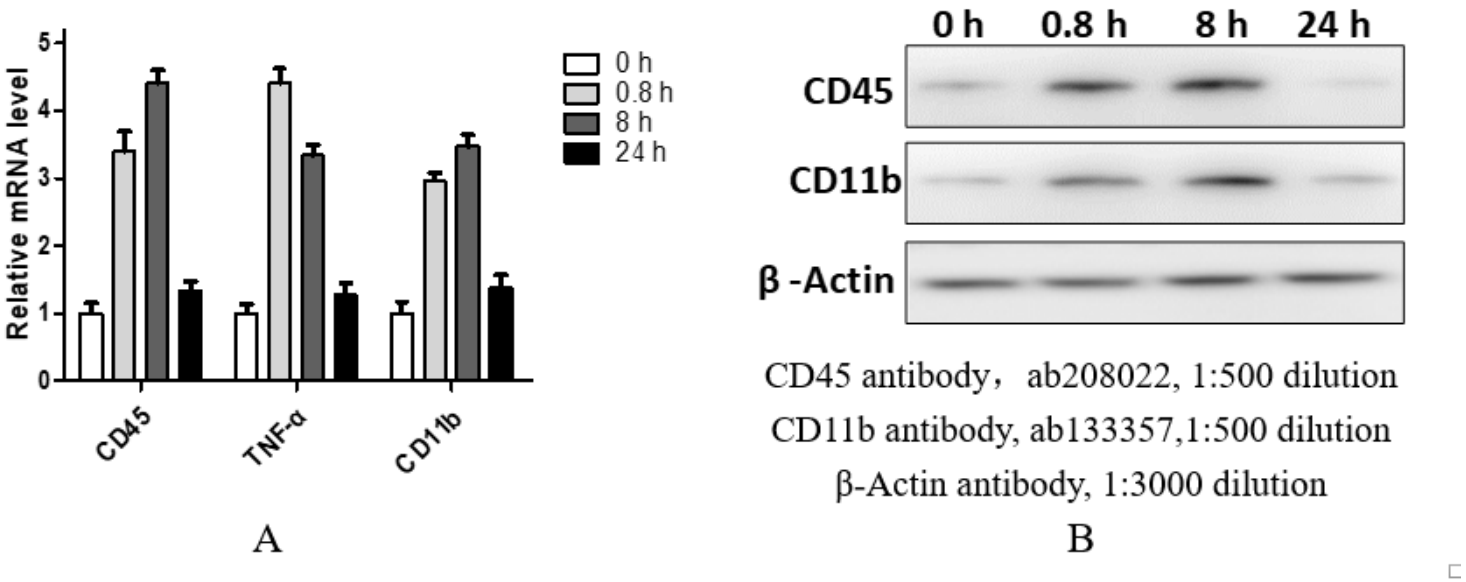

Figure 2

The expressions of M1 markers in microglial cells of mice with heat sickness, with the relative expressions of M1 markers in microglia using western blotting. A: Detection of the expressions of mouse microglia M1 type markers CD45, TNF-α, and CD11-b mRNA in different groups. The expressions of CD45 and CD11-b in the HS 8 group were significantly higher than those in the normal group, while those of CD45 and CD11-b in the HS 24 group decreased significantly when compared with the HS 8 group. B: Western blot analysis of the expressions of the four groups.

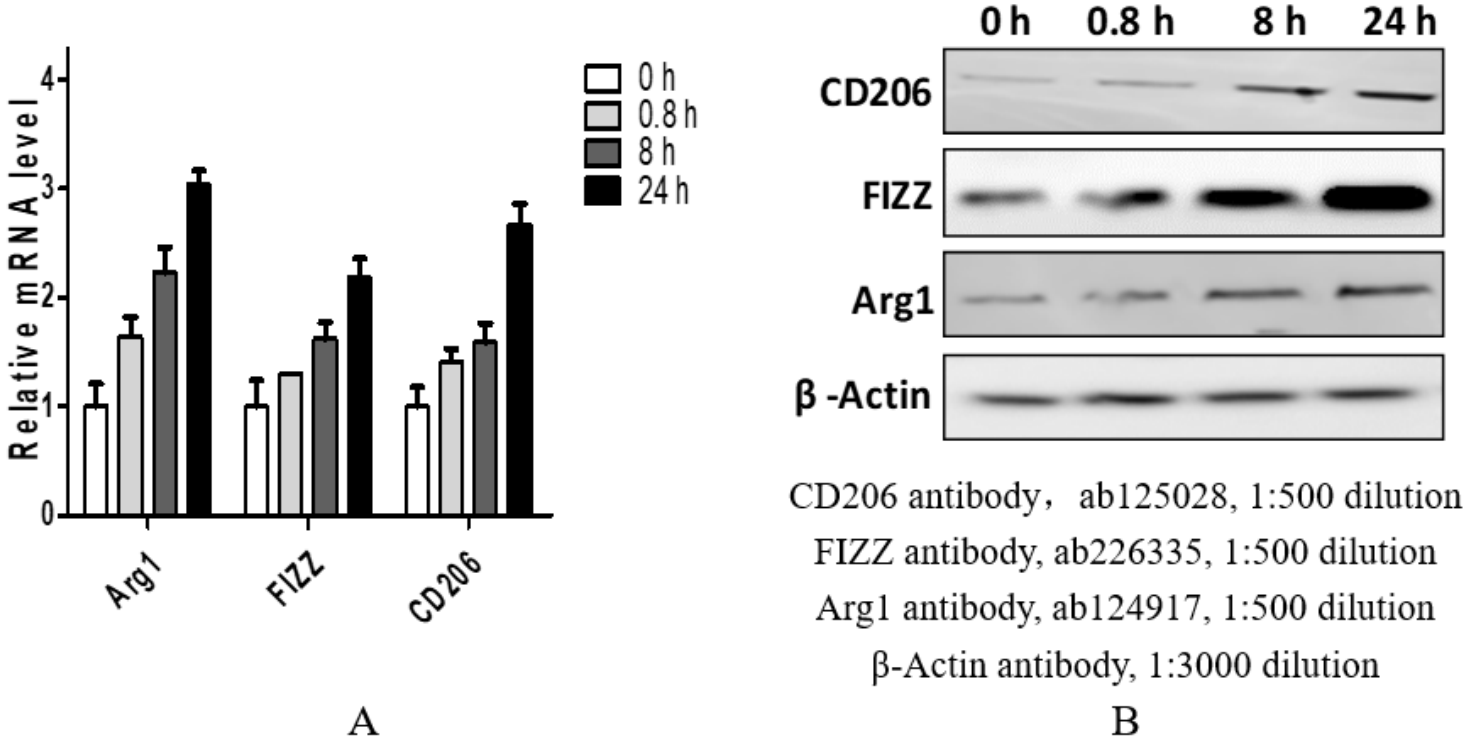

Figure 3

Expressions of M2 markers in microglial cells of mice with heat sickness; the relative expressions of M2 markers in microglia using western blotting. A: Detection of the expressions of mouse microglia M2 type markers CD206, FIZZ, and Arg1 mRNA in different groups. The expressions of CD206, FIZZ, and Arg1 in the HS 8 group began to rise when compared with the normal group, while expressions of CD206, FIZZ, and Arg1 were significantly higher than the normal group. B: Western blot analysis of the expressions of the four groups.

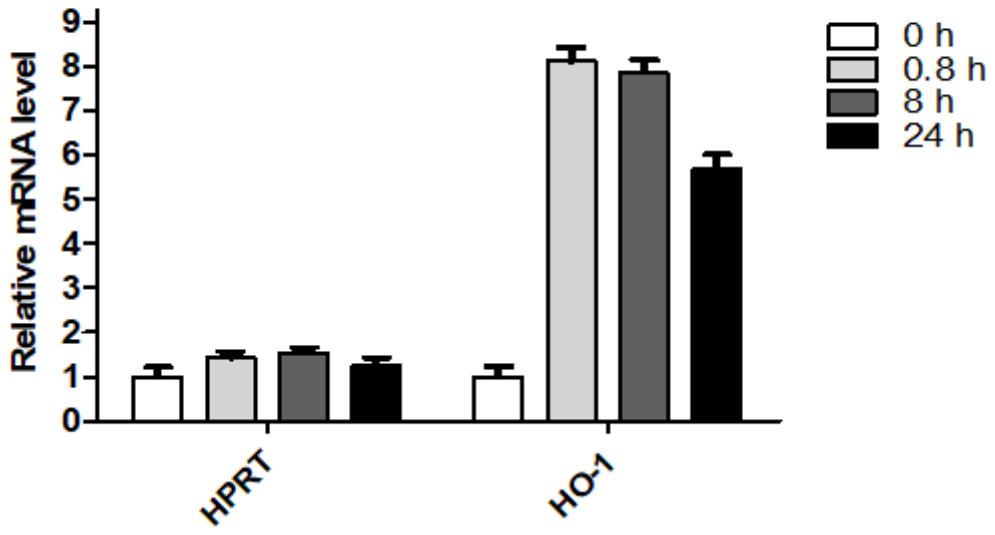

Figure 4

Expression of HPRT and HO-1 mRNA in microglia of brain tissue of mice with heat radiation disease. After heat stimulation, the mRNA expressions of HPRT and HO-1 genes in microglia of the four groups of mice. The HPRT expressions in each group of heat radiation disease showed no significant difference, while the HO 0.8 and HS 8 groups showed significantly higher HO-1 expressions than the normal group. The higher HO-1 expression in the HS 24 group was significantly lower than that in the normal group.

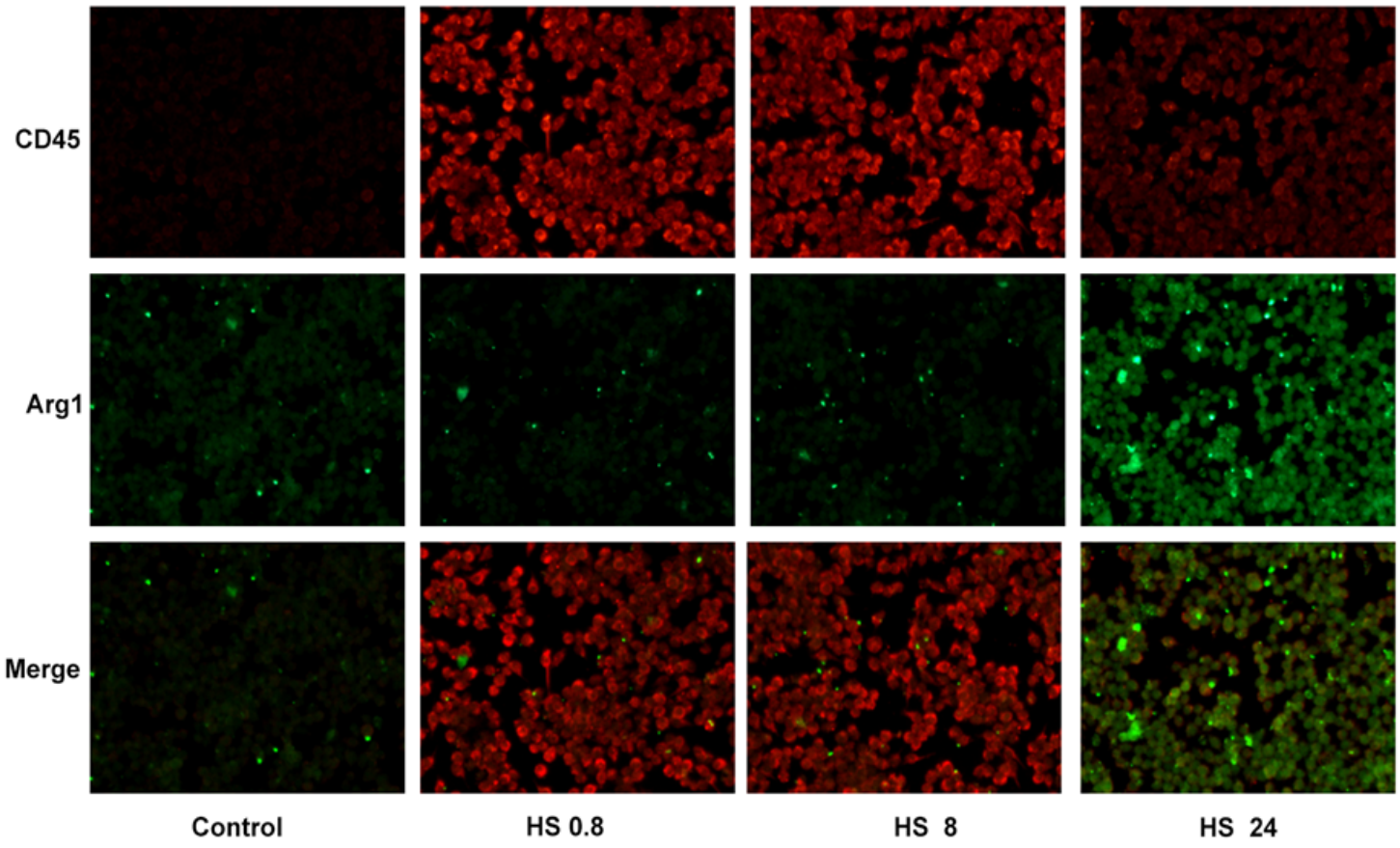

Figure 5

Fluorescence co-localization of microglial marker expression using immunohistochemistry (100 m). CD45 showed a red fluorescent label, and Arg1 showed a blue fluorescent label. The expression of CD45 began to be obvious with HS 0.8 expression, and HS 8 was significantly expressed when compared to the control group, while HS24 expression decreased and Arg1 significantly increased HS24 expression after heat injury.

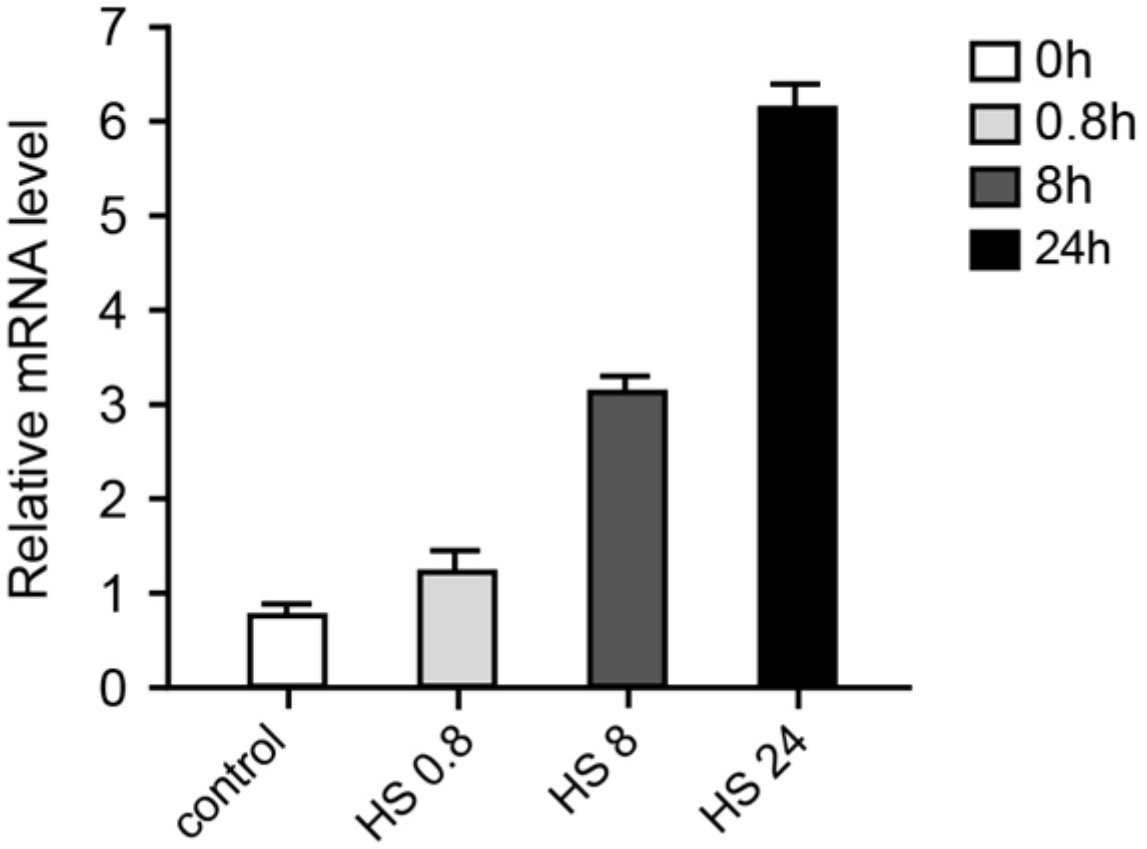

Figure 6

Circhipk3 expression in microglial cells. The qPCR was used to detect the expression of circHIPK 3 of microglia in the normal group and fever groups. The expressions of circhipk3 in microglia of the HS 0.8 and HS 8 groups were gradually increased compared with the normal group. The expression of the HS 24 group was significantly higher than that of the control group

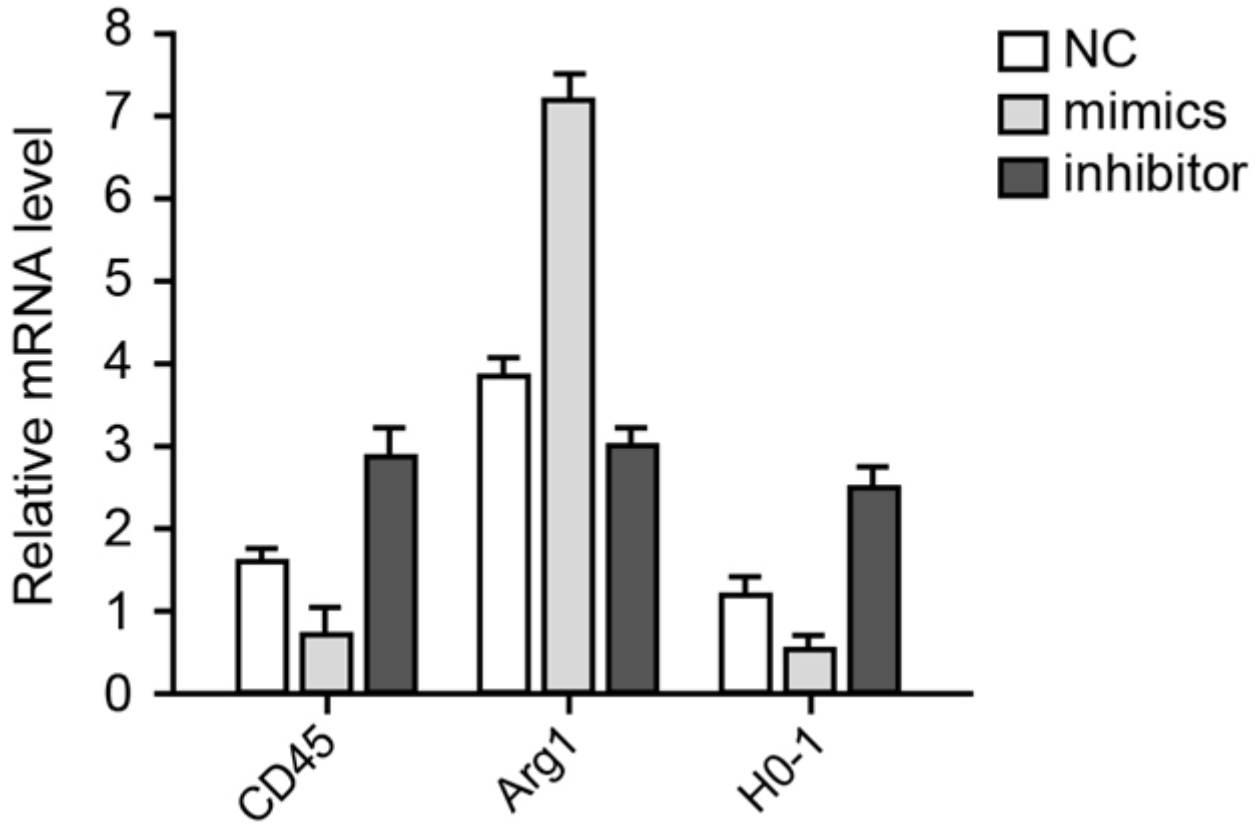

Figure 7

The circhipk3 participates in microglial polarization of heat radiation disease. The qPCR detected the expressions of M1 (CD45), M2 (Arg1), and HO-1 cytokines. Compared with the NC transfection group, the circhipk3 mimic significantly increased Arg1 expression and inhibited the expressions of CD45 and HO-1; while the circhipk3 inhibitor promoted the expressions of CD45 and HO-1 and suppressed the expression of Arg1.