Ⅱ-69

Modulatory effect of *Pueraria lobata* on the functional activation of macrophages induced by lipopolysaccharide

[a] School of Bioscience and Biotechnology, Kangwon National University
[b] Department of biochemistry, Kangwon National University
Ting Shen[a], Tao Yu[a], Se Eun Byeon[a], Yong Gyu Lee[a], Jong Dai Kim[a], Tae Woong Kim [b], and Jae Youl Cho[a, *]

Objectives

Pueraria lobata (Willd.) Ohwi was examined to evaluate its modulatory effects on the functional activation of macrophages under LPS treatment. To do this, nitric oxide (NO) production, reactive oxygen species (ROS) generation, cytoprotection and phagocytosis were selected to be tested using methanol extract from P. lobata (Pl-ME).

Materials and Methods

○ Materials

the total methanol extract of *Pueraria lobata* (Pl-ME) was identified by general method. DHR123 (Dihydrorhodamine123), sodium nitroprusside (SNP), FITC-dextran, MTT (3-[4,5-dimethylthiazol-2-yl]-2,5-diphinyltetrazolium bromide), and lipopolysaccharide (LPS, E. coli 0111:B4) were purchased from Sigma (St. Louis, MO). LY294002, wortmannin, U0126, SP600125, and SB203580 were obtained from Calbiochem (La Jolla, CA). Fetal bovine serum (FBS) was obtained from Hyclone (Hyclone, South Logan, UT, USA). All other chemicals were Sigma grade.

○ Methods

To do these experiments, anti-inflammatory effects such as nitric oxide (NO) production, MTT assay, Luciferase Reporter Gene Activity Assay, phagocytosis, ROS determination and RT-PCR were tested according to previous methods using murine macrophage cell line and Human embryonic kidney cell line.

Results

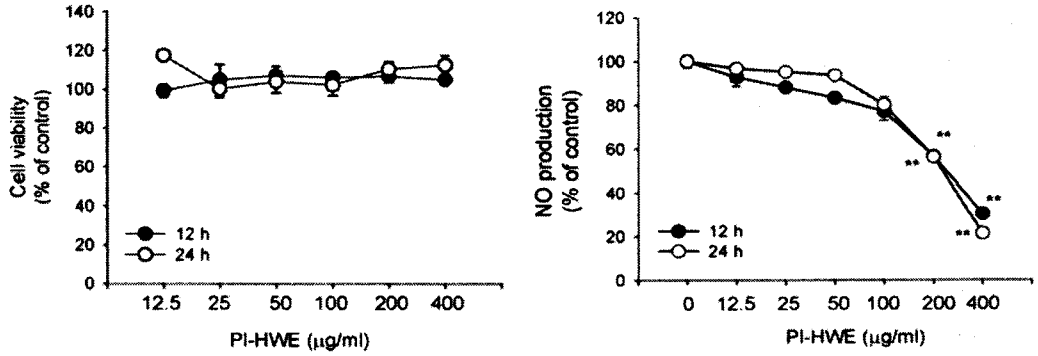
Pl-ME dose-dependently blocked NO production in LPS-stimulated RAW264.7 cells but not sodium prusside (SNP)-generated NO release. The NO inhibition seemed to be due to blocking inducible NO synthase (iNOS), since PI-ME suppressed its expression in a NF-κ-independent manner. In agreement, this extract also effectively protected RAW264.7 cells from LPS-induced cytotoxicity. However, PI-ME did not block ROS generation and rather it enhanced. Finally, this extract negatively modulated FITC-dextran uptake. Therefore, our data suggested that PI-ME may be involved in negatively regulating some macrophage-mediated inflammatory responses such as NO production and phagocytic uptake.

Corresponding autho : 조재열 E-mail : jaecho@kangwon.ac.kr Tel : 033-250-6488

This work was (in part) supported by the Minstry of Commerce, Industry and Energy through the Center for Efficacy Assessment and Development of Functional Foods and Drugs at Hallym University, Korea.

Fig.1 Cell viability and LPS-induced NO production of PI-HWE in RAW264.7 cells in a dose-dependent manner.

(A) Cell viability (B) NO production



(C) mRNA expression level of iNOS

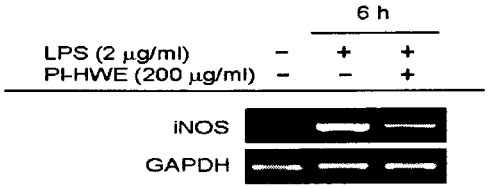


Fig.2 Phagocytotic uptake of PI-HWE in RAW264.7 cells.