S3-2

Immuno-Modulating Effects and Anti-tumor Activities of Proteoglycan Isolated from *Phellinus linteus*: Phenotypic and Functional Maturation of Murine Dendritic Cells *in vitro* and *in vivo*

Jae-Dong Lee[1], Gi-Young Kim[2], and Yeong-Min Park[2]

[1]Department of Microbiology, College of Natural Sciences, Jangjeong-dong, Keumjeonggu, Pusan National University, Pusan, Korea; [2]Department of Microbiology, College of Medicine, Pusan National University, Ami-dong 1-10, Seogu, Pusan, Korea

Proteoglycan (PG) isolated from *Phellinus linteus* are known to stimulate the proliferation of T lymphocytes and humoral immune functions to act as a polyclonal activator of B cells, and to inhibit tumor growth and metastasis. However, little is known about its immunomodulating effects or the effects of its mechanisms on murine bone marrow (BM)-derived dendritic cells (DC). In this study, it profoundly increased CD80, CD86, MHC I, and MHC II expression in murine, GM-CSF and IL-4 stimulated, DC. The ability of unstimulated DC to uptake dextran was higher than that of PG- or LPS-stimulated DC. The production of IL-12 from DC was analyzed using flow cytometry and ELISA. Untreated DC secreted a low concentration of IL-12, while PG- or LPS-stimulated DC secreted higher levels of IL-12 than untreated DC. There were no remarkable differences in the concentrations of IL-12 produced by PG- or LPS-stimulated DC. PG-treated DC were much more potent antigen-presenting cells in allogeneic reactions than untreated DC. PG treatment not only formed morphologically mature DC but also induced predominant migration to lymphoid tissues. Moreover, the inhibitors of protein tyrosine kinase (PTK) or protein kinase C (PKC) significantly blocked the expression of surface molecules and IL-12 production in PG-stimulated DC. Treatment of DC with PG directly induced PKC activity and phosporylated PTK.

Administration of *in vivo* PG strongly inhibited the MCA-102 tumor growth and volume through immuno-stimulating activities. When the ratio of $CD8\alpha^+$ DC to $CD8\alpha^-$ DC increased, PG enhanced IL-12 and IFN-γ production, and the expression of surface molecules including major histocompatibility complexes (MHC) classes I and II, as well as CD80 and CD86. PG also caused a marked increase of production of Th-1 cytokine (IFN-γ) and decrease of production of Th-2 cytokine (IL-4) by splenic cells and inguinal lymph node cells in MCA-102 tumor-bearing mice. Furthermore, PG stimulated the proliferation of $CD4^+$ and $CD8^+$ T cells. In addition, examination of the tumor rejection effects of combination therapy of PG-based chemotherapy and tumor lysate-pulsed (TP) DC-based immunotherapy completely inhibited the growth of MCA-102 cells. These results indicate that the administration of PG inhibited the tumor growth of MCA-102 through a mechanism leading to a Th-1 dominant immune state and activation of $CD11c^+CD8\alpha^+$ DC. Thus, PG may be useful for the prevention of cancer growth.