HEPATOLOGY
Official Journal of the American Association for the Study of Liver Diseases

Galectin-3 Deficiency Prevents Concanavalin A–Induced Hepatitis in Mice

Vladislav Volarevic,[1] Marija Milovanovic,[1] Biljana Ljujic,[1] Nada Pejnovic,[1] Nebojsa Arsenijevic,[1] Ulf Nilsson,[3] Hakon Leffler,[4] and Miodrag L. Lukic[1]

We used concanavalin A (Con A)-induced liver injury to study the role of galectin-3 (Gal-3) in the induction of inflammatory pathology and hepatocellular damage. We tested susceptibility to Con A–induced hepatitis in galectin-3-deficient (Gal-$3^{-/-}$) mice and analyzed the effects of pretreatment with a selective inhibitor of Gal-3 (TD139) in wild-type (WT) C57BL/6 mice, as evaluated by a liver enzyme test, quantitative histology, mononuclear cell (MNC) infiltration, cytokine production, intracellular staining of immune cells, and percentage of apoptotic MNCs in the liver. Gal-$3^{-/-}$ mice were less sensitive to Con A–induced hepatitis and had a significantly lower number of activated lymphoid and dendritic cells (DCs) in the liver. The level of tumor necrosis factor alpha (TNFα), interferon gamma (IFNγ), and interleukin (IL)-17 and -4 in the sera and the number of TNFα-, IFNγ-, and IL-17- and -4-producing cluster of differentiation (CD)4^+ cells as well as IL-12-producing $CD11c^+$ DCs were lower, whereas the number of IL-10-producing $CD4^+$ T cells and $F4/80^+$ macrophages were significantly higher in livers of Gal-$3^{-/-}$ mice. Significantly higher percentages of late apoptotic Annexin V^+ propidium-idodide$^+$ liver-infiltrating MNCs and splenocytes were observed in Gal-$3^{-/-}$ mice, compared to WT mice. Pretreatment of WT C57BL/6 mice with TD139 led to the attenuation of liver injury and milder infiltration of IFNγ- and IL-17- and -4-producing $CD4^+$ T cells, as well as an increase in the total number of IL-10-producing $CD4^+$ T cells and $F4/80^+$ $CD206^+$ alternatively activated macrophages and prevented the apoptosis of liver-infiltrating MNCs. *Conclusions*: Gal-3 plays an important proinflammatory role in Con A–induced hepatitis by promoting the activation of T lymphocytes and natural killer T cells, maturation of DCs, secretion of proinflammatory cytokines, down-regulation of M2 macrophage polarization, and apoptosis of MNCs in the liver. (HEPATOLOGY 2012;55:1954-1964)

Concanavalin A (Con A)-induced liver injury is a well-established murine model of T-cell-mediated hepatitis. Intravenous (IV) injection of Con A induces acute liver injury and systemic immune activation in mice that resembles the pathology of immune-mediated hepatitis in humans.[1] Activated T cells have a critical role in Con A–induced liver damage.[1] Cluster of differentiation (CD)4^+ T lymphocytes infiltrate the liver tissue and secrete large amounts of cytokines, such as tumor necrosis factor alpha (TNFα), interferon gamma (IFNγ), interleukin (IL)-2 and -6, and granulocyte macrophage colony-stimulating factor.[1,2] Apart from cluster of differentiation (CD)4^+ T cells, $CD8^+$ T cells, natural killer (NK), natural killer T (NKT) cells, and macrophages could induce hepatocyte cell death by either cell-to-cell contact, through the secretion of proinflammatory cytokines, or reactive oxygen species.[1-4]

Abbreviations: ALEs, advanced lipoxidation endproducts; ALT, alanine aminotransaminase; APC, allophycocyanin; AST, asparate aminotransferase; CD, cluster of differentiation; Con A, concanavalin A; DCs, dendritic cells; ELISA, enzyme-linked immunosorbent assay; FITC, fluorescein isothiocyanate; Foxp3, forkhead box protein 3; Gal-3, galectin-3; Gal-$3^{-/-}$, Gal-3 deficient; Gal-3-INH, Gal-3 inhibitor; HBV, hepatitis B virus; IFNγ, interferon gamma; IL, interleukin; IP, intraperitoneal; IV, intravenous; MNC, mononuclear cell; NASH, nonalcoholic steatohepatitis; NK, natural killer; NKT, natural killer T; PE, phycoerythrin; PI, propidium idodide; SEM, standard error of the mean; TD139, selective inhibitor of Gal-3; Th, T-helper cells; TNFα, tumor necrosis factor alpha; Tregs, T regulatory cells; WT, wild type.

From the [1]Center for Molecular Medicine and Stem Cell Research, Faculty of Medicine, University of Kragujevac, Kragujevac, Serbia; [2]Organic Chemistry, Lund University, Lund, Sweden; and [3]Section MIG, Department of Laboratory Medicine, Lund University, Lund, Sweden.

Received August 19, 2011; accepted December 11, 2011.

This study was supported by grants (ON175069, ON175071, and ON175103) from the Ministry of Science and Technology, Republic of Serbia.

Address reprint requests to: Miodrag L. Lukic, M.D., Ph.D., Center for Molecular Medicine and Stem Cell Research, Faculty of Medicine, University of Kragujevac, 69 Svetozara Markovica Street, 34000 Kragujevac, Serbia. E-mail: miodrag.lukic@medf.kg.ac.rs; fax: +38134306800 ext. 112.