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The Role of Tumor Necrosis Factor Receptor (TNFR) Superfamily Proteins in *Listeria monocytogenes* Infection

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The interactions between members of the tumor necrosis factor (TNF) family and their specific receptors (TNFRs) play critical roles in controlling immune cell division, life, and death. However, little is known of its role in bacterial infections. Recently, we have found that 4-1BB (CD137) and herpes virus entry mediator (HVEM) molecules exert inflammatory responses and killing of bacteria *in vitro* and *in vivo* animal model.

4-1BB-deficient (4-1BB$^{-/-}$) mice were much more susceptible to *Listeria monocytogenes* (LM) infections than wild-type mice. Upon *L. monocytogenes* infection, 4-1BB$^{-/-}$ mice showed a lower survival rate, a higher bacterial burden in organs, and larger hepatic microabscesses than wild type mice. 4-1BB$^{-/-}$ mice also had impairment in clearance of bacteria from the bloodstream. Neutrophils from wild type mice constitutively expressed 4-1BB, which could be activated to induce intracellular Ca^{2+} influx by ligation with anti-4-1BB antibody. On the other hand, neutrophils from 4-1BB$^{-/-}$ mice were defective in reactive oxygen species generation, phagocytic activities, and intracellular Ca^{2+} mobilization. In addition, mice pretreated with anti-4-1BB antibody were much more resistant to *L. monocytogenes* infection than control antibody-treated mice. Administration of an agonistic anti-4-1BB monoclonal antibody (3E1), led to rapid infiltration of CD11b^{+}Gr1^{hi} and CD11b^{+}Gr1^{int} cells, identified as neutrophils and monocytes respectively, into the LM-infected livers. In the 3E1-treated mice, serum levels of chemokines and cytokines responsible for recruitment and activation of neutrophils and monocytes increased as early as 30 min post infection and peaked by 2 h, while those in control antibody-treated mice only started to increase 16 h post infection. The monocytes and neutrophils from the 3E1-treated mice had higher levels of activation markers, phagocytic activity and reactive oxygen species than those from control mice. *In vitro* stimulation of neutrophils with 4-1BB induced the production of IL-6, TNF-α and IL-10 and enhanced their phagocytic activities. Our results suggest that 4-1BB stimulation in the early phase of LM infection plays a critical role in eliminating the infection.

HVEM is highly expressed in monocytes and neutrophils, and its interaction with its specific ligand "homologous to lymphotoxins, shows inducible expression, and competes with herpes simplex virus glycoprotein D for HVEM/tumor necrosis factor (TNF)-related 2" (LIGHT) enhanced bactericidal activity against *Listeria monocytogenes* and *Staphylococcus aureus*. The LIGHT-HVEM interaction increased levels of phagocytosis, interleukin (IL)-8, TNF-α, nitric oxide (NO), and reactive oxygen species (ROS) in monocytes and neutrophils. Anti-HVEM monoclonal antibody was able to block LIGHT-induced bactericidal activity, cytokine production (IL-8 and TNF-α), and ROS generation. Inhibition of ROS and NO production blocked LIGHT-induced bactericidal activity. In addition, we found that LIGHT induces rapid elevation of intracellular Ca^{2+} in a HVEM-specific manner in parallel with TNF-α production, and enhances the bactericidal activities of monocytes. Immunoprecipitation and Western blotting analyses revealed phosphorylation of PLCγ1 but not PLCγ2. LIGHT-induced Ca^{2+} response was completely abolished by silencing PLCγ1, or preincubating monocytes with PLC inhibitors, antagonists of the inositol-1,4,5-triphosphate receptor, or Ca^{2+} chelators. Furthermore, these PLC/Ca^{2+} inhibitors also blocked rhLIGHT-mediated IκBα degradation, generation of reactive oxygen species, TNF-α production and the bactericidal activities of monocytes.

References

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