

Mouse model of human hantavirus infection

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Hantaviruses cause hemorrhagic fever with renal syndrome (HFRS) and hantavirus cardiopulmonary syndrome (HCPS) in humans after transmission from chronically infected but asymptomatic rodents, the natural reservoir hosts (4). In contrast to rodents, humans eliminate hantaviruses (immunological species barrier). The hallmarks of hantavirus-associated pathogenesis in humans are increased vascular permeability and loss of platelets (acute thrombocytopenia). Intriguingly, hantaviruses do not cause any direct cytopathic effect suggesting that an overstimulated human immune response is responsible for both the immunological species barrier and the symptoms observed *in vivo* (4). In line with this view, we have demonstrated that pathogenic hantaviruses infect dendritic cells (DC), the master regulators of the antiviral immune defence, which subsequently mature and acquire strong T cell-stimulatory capacity (3). Moreover, our experiments revealed that after infection with pathogenic hantaviruses, monocytes survive and develop into DC-like cells (4). Analysis of hantavirus-induced immunopathogenesis is hampered by the fact that no suitable animal model is available so far and normal laboratory mouse strains are not susceptible to hantavirus infection. Therefore, we will use humanized mice to test the concept that the immunological species barrier for hantaviruses in humans is inevitably linked to hantavirus-induced immunopathogenesis *in vivo*.

We have demonstrated *in vitro* that pathogenic but not apathogenic hantaviruses infect human megakaryocytes (2) which upon maturation generate platelets. Intriguingly, despite of high level hantavirus replication megakaryocytic cell differentiation was not disturbed (2). However, strong upregulation of HLA class I molecules occurred supporting the concept that pathogenic hantaviruses interfere with megakaryopoiesis *in vivo* through induction of immune mechanisms (2).

In the first step we will establish humanized mice that permit long term human megakaryopoiesis and human immune responses. For this purpose irradiated nonobese diabetic/severe combined immunodeficiency interleukin-2 receptor γ -chain knockout (NOD-*scid IL2r γ ^{null}*) mice expressing human HLA-A2/human β 2M will be transplanted with human CD34+ cells. Viral replication after hantavirus infection will be assessed in blood and organs (bone marrow, spleen, kidney, liver, heart) by quantitative real time PCR. Furthermore, hantavirus-specific immune responses of human T and B cells will be analyzed (ELISA, cytokine secretion and cytotoxicity assays). Moreover, platelet counts will be determined after hantavirus infection to verify the significance of the mouse model. In a second step the pathogenetic mechanisms underlying acute thrombocytopenia will be analyzed. To this end it will be determined whether the immature platelet fraction (IPF) is decreased indicating disturbed megakaryopoiesis. It will be investigated whether the human megakaryocytes in the bone marrow are eliminated. To test the concept of immunopathogenesis immune cells such as cytotoxic T cells will be depleted in the mouse model before infection and platelet production will be assessed. Finally, the disease-inducing capacity of different hantavirus strains will be compared. In a third step (outlook) experimental vaccines such as virus-like particles and candidate drugs will be tested in humanized mice.

Literature:

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