The role of pro- and anti-apoptotic proteins for homeostasis and function of mast cells

Mast cells exert beneficial as well as detrimental functions in host defense and various diseases, but the regulation of mast cell homeostasis is only partially understood. Mcl-1 and Bcl-xL belong to the group of anti-apoptotic Bcl-2 proteins and play essential roles in embryogenesis and survival of many cell types. Tumor necrosis factor-related apoptosis-inducing ligand receptors (TRAIL-Rs) participate in the death receptor pathway of apoptosis. In the present study, we aimed to investigate the role of Mcl-1, Bcl-xL and TRAIL-R in mast cells *in vivo*.

We generated mouse models with mast cell-specific deletion of *Mcl-1*, *Bcl-x* and *TRAIL-R* by crossing *Mcl-1*$^{fl/fl}$, *Bcl-x*$^{fl/fl}$ and *TRAIL-R*$^{fl/fl}$ mice to the *Mcpt5Cre* strain, which expresses Cre recombinase selectively in connective tissue type mast cells. For *in vitro* experiments, we crossbred *Bcl-x*$^{fl/fl}$ with *Mx1Cre* and *TRAIL-R*$^{fl/fl}$ with *DelCre* mice.

Mast cell-specific deletion of *Mcl-1* resulted in ablation of mast cells in various tissues. Numbers of T cells, B cells, dendritic cells, macrophages and granulocytes were not altered, confirming that depletion of mast cells is highly specific in this mouse model. Furthermore, *Mcpt5Cre/Mcl-1*$^{fl/fl}$ mice were completely protected from IgE-induced passive systemic anaphylaxis, which is mediated by mast cells. *Mcpt5Cre/Bcl-x*$^{fl/fl}$ mice exhibited only a partial decrease of mast cell counts and IgE-mediated anaphylaxis was not affected. Interestingly, deletion of *Bcl-x in vitro* resulted in increased levels of Mcl-1. Also, *Bcl-x*-deficient mast cells showed enhanced apoptosis in response to actinomycin D, etoposide and TRAIL in conjunction with actinomycin D. The characterization of *Mcpt5Cre/TRAIL-R*$^{fl/fl}$ mice revealed normal numbers of mast cells and a slight increase in the anaphylactic response. Of note, activation of KIT by SCF or activating mutations induced TRAIL-mediated apoptosis in mast cells.

Collectively, the present study defines for the first time differential roles of Mcl-1 and Bcl-xL in homeostasis of mast cells. The *Mcpt5Cre/Mcl-1*$^{fl/fl}$ cross represents a novel mouse model of mast cell deficiency, which will be highly useful for analyzing functions of connective tissue type mast cells. Our data also show that KIT activation regulates the function of TRAIL-Rs, which may have clinical implications for the treatment of mast cell-associated diseases.