# Developmental and Molecular Immunology

**Antonius Rolink**
Department of Biomedicine
Roche Professor of Immunology
University of Basel

## Group Members

Llucia Ines Alberti Servera (PhD Student)
Sofia Alves (Technician)
Melissa Amrein* (Undergraduate Student)
Prof. Dr. Jan Andersson (Research Associate)
Ilda Blanco-Alves Vaz (Technician)
Giuseppina Capoferri (Technician)
Grozdan Cvijetic (Undergraduate Student)
Marianne Dölz* (Undergraduate Student)
Corinne Engdahl Démollière (Technician)
Nadine Gehre* (PhD Student)
Stefan Heiler (PhD Student)
Fabian Klein (PhD Student)
Jonas Lötscher (MD-PhD Student)
Mladen Mitrovic (Undergraduate Student)
Dr. Natko Nuber* (Postdoc)
Julia Obenauer* (Undergraduate Student)
Petra Pfenninger* (Undergraduate Student)
Johanna Rolink (Technician)
Michael Rolink (Technician)
Nicole Salvisberg (Administrative Assistant)
Dr. Panagiotis Tsapogas (Postdoc)
Audrey Lilly von Münchow (Postdoc)
Ernst Wagner* (Research Associate)
Oliver Wirz* (Undergraduate Student)
Simon Zinkhan* (Undergraduate Student)

*left during report period

## Molecular mechanisms guiding hematopoietic cell development

**A permissive role for IL-7 in mouse B cell development**

Patients deficient for IL-7 or the IL-7R are practically devoid of T cells but have normal B cell compartments. In mice deficient for IL-7 or the IL-7R both T cell and B cell compartments are dramatically reduced. Based on these findings it was argued that IL-7 plays a redundant role in human B cell development but a non-redundant and may be even an instructive role in mouse B cell development. Now we challenged this hypothesis by analyzing the by us generated FLT3L transgenic mice. These transgenic mice posses dramatically increased populations of progenitor cells in the bone marrow including the pre-pro-B cells. Now by generating IL-7 deficient mice on a FLT3L transgenic background we found a very significant rescue of all the precursor B cell subpopulations in the bone marrow and the mature B cell populations in the periphery. Thus, increased levels of FLT3L make the role of IL-7 in mouse B cell development largely redundant. Our findings rather suggest that IL-7 mainly acts as a growth factor for pre-B cells and thereby making B cell development more efficient. Yet, another suggestion coming from our studies is that human B cell development is regulated by FLT3L.

**Hematopoietic progenitors with multipotent developmental potential: real multipotency or heterogeneity?**

Over the past couple of years new hematopoietic progenitors with multipotent developmental potential mainly determined *in vitro* were described. However, by the generation of so-called reporter mice the multipotency of some of these precursors *in vivo* was questioned. Moreover, the multipotent developmental potential *in vitro* of these precursors was tested at the "bulk" but not at the single cell level. Therefore, the *in vivo* significance of this multipotency is questionable and furthermore the *in vitro* experiments do not exclude heterogeneity within the tested precursor populations.

Several years ago we have described a small bone marrow derived precursor population with lymphoid and myeloid developmental potential *in vitro* (Balciunaite G, Ceredig R, Massa S, Rolink AG. Eur J Immunol. 2005 Jul;35(7):2019-30). This population comprises 0.1 – 0.3% of total BM nucleated cells and was characterized as being B220+ c-Kit+ CD19- NK1.1-. Now, by identifying new markers and by analyzing newly generated mutant mice we have found that this population can be subdivided into at least 6 subpopulations. The main marker we used for this was Ly6D. Single cell RNA- sequence analysis performed on the Ly6D+ and Ly6D-, B220+ c-Kit+ CD19- NK1.1- cells revealed within the Ly6D+ cells a largely lymphoid restricted transcriptome whereas the Ly6D- cells showed either a transcriptome reminiscent of lymphoid, myeloid or dendritic cells. Thus these findings reveal that B220+ c-Kit+ CD19- NK1.1- cells are very heterogeneous and that myeloid and lymphoid developmental potential is coming from different cells within this population.

## Selected Publications

Gehre N, Nusser A, von Muenchow L, Tussiwand R, Engdahl C, Capoferri G, Bosco N, Ceredig R, Rolink AG. (2015) A stromal cell free culture system generates mouse pro-T cells that can reconstitute T- cell compartments *in vivo*. European journal of immunology 45, 932–942

Nusser A, Nuber N, Wirz OF, Rolink H, Andersson J, Rolink A. (2014) The development of autoimmune features in aging mice is closely associated with alterations of the peripheral CD4(+) T-cell compartment. European journal of immunolo- gy 44, 2893–2902

Swee LK, Nusser A, Curti M, Kreuzaler M, Rolink H, Terracciano L, Melchers F, Andersson J, Rolink A. (2014) The amount of self-antigen determines the effector function of murine T cells escaping negative selection. European journal of immunology 44, 1299–1312

Tsapogas P, Swee LK, Nusser A, Nuber N, Kreuzaler M, Capoferri G, Rolink H, Ceredig R, Rolink A. (2014) *In vivo* evidence for an instructive role of fms-like tyrosine kinase-3 (FLT3) ligand in hematopoietic development. Haematologica 99, 638–646

von Muenchow L, Engdahl C, Karjalainen K, Rolink AG. (2014). The selection of mature B cells is critically dependent on the expression level of the co-receptor CD19. Immunology letters 160, 113–119

## Connection to Clinical Practice

**Prof. Dr. Antonius Rolink**
University of Basel, Department of Biomedicine

**The prophylactic and/or therapeutic effects of IL-2 – anti - IL-2 complexes on SLE like cGVHD**

cGVHD as can be induced by the injection of DBA/2 T cells into semi-allogeneic (C57/Bl6 x DBA/2) F1 (BDF1) mice shows many similarities to SLE in man. Some years ago it was shown that injection of IL-2 anti-IL2 complexes into normal mice can result into a dramatic increase of regulatory T cells (Tregs) or CD8 T cells. The increase of either Tregs or CD8 T cells was dependent on the anti-IL2 antibody that was used for generating the complexes. We have now tested the prophylactic and therapeutic effects of these complexes on SLE like cGVHD. Our findings indicate that the treatment of BDF1 mice before the injection of DBA/2 T cells with complexes that induce a Treg increase to a large extent prevents the development of cGVHD whereas the injection of the complexes that induce a CD8 increase had no effect or even made the disease worse.

In marked contrast no amelioration of the cGVHD was observed when IL-2 complexes that induce Treg cells were given 3 weeks after induction of a cGVHD (therapeutic protocol). However, the therapeutic treatment that induces a CD8 increase very significantly ameliorated the autoimmunity as determined by the titer of autoantibodies and the development of immune complex glomerulonephritis. No improvement of disease by these complexes was observed when the cGVHD was induced by CD8 depleted DBA/2 T cells. This finding suggest that the ameliorating effects by the therapeutic treatment with CD8 inducing complexes might be due to an increase of DBA/2 CD8 T cells.

Overall, these studies indicate that IL-2 complexes might well be envisaged for prophylactic and/or therapeutic therapies of cGVHD and may be even SLE.