IP Internship project 5:
Antibodies in movement: exchange of antigen binding sites (half-molecules) in IgG4

Introduction: Only 3-4% of human IgG consists of IgG4. However, due to prolonged exposure to antigen (such as beekeepers with bee venom) more than 90% of the specific antibody response may consist of IgG4. IgG4 is considered to be a 'blocking' antibody: it does bind antigen, but does not subsequently activate the immune system. IgG4 can therefore play an important role in the development of tolerance

One of the features that makes IgG4 so special, is the possibility that half of an antibody interacts with half of another antibody. As a result an antibody with two different antigen binding sites is developed. Because the process takes place at random, any specificity is combined with another random specificity *in vivo*. So effectively, these antibodies are monovalent, which prevents development of immune complexes by cross-linking. Under the right circumstances, this process may also occur *in vitro*. The mechanism of this process is not well known.

Aim: The goal of this project is to find out how the exchange process works. This will include labeling IgG4 antibodies with fluorescent groups and the measuring the speed of exchange. The speed of the reaction can be related to the structure of the IgG antibody (wild type versus mutant) and the reaction conditions (concentration, temperature). Therefore, it might help with elucidating the mechanism. These measurements will be complemented by characterization of the bispecific antibody that is formed.

Techniques
- Fluorescence spectroscopy/FRET
- Reaction kinetics
- SDS-PAGE
- RIA
- HP-SEC

Duration: at least 5 months. Students that study biochemistry, biophysical chemistry or a related study, who are looking for a dynamic and interesting internship and are interested in the above project are encouraged to contact the group leader, Theo Rispens, either by e-mail: t.rispens@sanquin.nl.