

## IDS TCW QUARTERLY NEWLETTER – 2/2009

[www.immunologyofdiabetessociety.com/committee\\_t-cell.htm](http://www.immunologyofdiabetessociety.com/committee_t-cell.htm)

Sept 4, 2009

Dear Colleagues,

During IDS-10, organized by Ake Lernmark and Corrado Cilio in Malmö on May 17-20 2009, the TCW group presented the rationale and outline of the current projects.

Tim Tree introduced the project on PBMC freezing and thawing (IDS-TCW Freezing Study I), Ivana Durinovic-Bello and Roberto Mallone reported on the initial phase of the Tetramer Directed Epitope Validation Initiative (proto-TDEVI Study) and Alessandro Sette presented data on epitope databases around infectious diseases.

In addition, selected abstracts on T cell studies were presented by Bart Roep (Detection of Islet-Specific Autoreactive T-Cells by a Novel Robust and High Through-Put Screening Strategy in Stored Blood Samples of Type 1 Diabetic), Stuart Mannering (The A-Chain of Insulin is a Hot-Spot for CD4+ T-Cell Epitopes in Human Type 1 Diabetes) and Howard Davidson (CD4 T Cell Autoreactivity to the Major Diabetes Autoantigen ZnT8).

Furthermore, we had our first “T Cell Karaoke” on one of the evenings in Malmö as an informal event where we openly discussed T cell issues and enjoyed good music and good company. For the next IDS, we thought of moving this nice event at an earlier evening hour, perhaps parallel to poster sessions.

Besides this meeting in Malmö, the group keeps interacting through monthly telephone conferences, initially by Skype, and now kindly supported by JDRF (Olivia Lou) with a telephone line conference of much better quality. During these meetings, we perform project planning and updating on a regular basis, and open issues are discussed among the group members. We are currently preparing a review manuscript to outline the background and rationale for current studies. Also, in press manuscripts regarding T cell projects are circulated amongst the group members. The chair of the telephone conference and the secretary (taking minutes) rotate every month. As announced previously, the TCW is focusing on three major projects:

**1) T1D T Cell Epitope Database:** starting from her recently published review article, Teresa DiLorenzo has updated and posted on our website a comprehensive database including both CD4+ and CD8+ epitopes, mouse and human. This database will be periodically updated, and you can notify omissions, errors or additions directly to the Author, as indicated on the website. Please note that data will be entered based exclusively on published reports, and scored according to different “degrees of evidence”, depending on results included in the quoted report.

**2) IDS TCW Freezing Study I:** the goal of this study is to compare two mainstream PBMC freezing/thawing protocols, namely cold vs. warm freezing medium (DMSO-human serum), along with other local protocols which may be considered as promising alternatives. The aim is to define starting “gold standard” freezing procedure(s) which perform superiorly across different T cell assay formats (i.e., best cell recovery and viability, preservation of beta-cell-specific T cell responses from frozen aliquots as compared to starting fresh PBMC preparations).

In an effort to blind the study, we elected to do so with antigens rather than with PBMC samples (see website for details). So far, antigens provided by each laboratory have been

coded by Tim Tree and sent back to the requesting laboratories. Two out of 9 labs already tested freshly isolated and frozen PBMCs from 5 patients with type 1 diabetes and 5 control subjects and thereby finished the first part of this study. The remaining labs are expected to finish within the next few months. All raw data will be sent to Tim Tree lab for de-coding and will be analysed by a group of people. Antigen identity will thus be unblinded only after data has been locked.

**3) Proto-TDEVI Study:** this is the first phase of a larger TDEVI (Tetramer-Directed Epitope Validation Initiative) study promoted by the Benaroya Research Institute in Seattle, under the sponsorship of JDRF and coordinated by Ivana Durinovic-Bello. The aim of TDEVI is to promote a strategy of epitope validation, based on independent, blinded tetramer testing performed in multiple Laboratories on a limited number of blood samples.

This first phase is ongoing, and includes a CD4+ and a CD8+ T cell arm involving the 9 Labs of the TCW Committee. It is expected to provide proof of principle about the feasibility of the strategy along with independent validation of a first limited set of epitopes: HLA-DR0401-restricted GAD<sub>270-285</sub>, GAD<sub>554-567</sub>, PPI<sub>76-90</sub>; and HLA-A2-restricted GAD<sub>114-122</sub>, GAD<sub>536-545</sub>, PPI<sub>34-42</sub> and PPI<sub>101-109</sub>. The required reagents and tetramers have been kindly provided by Dr. Jerry Nepom's laboratory and have been sent to participating laboratories. Detailed protocols have been distributed and lab personnel trained where necessary.

Three laboratories have started CD4+ Tetramer analyses and three more have commenced the CD8+ T cell arm. This phase of the study should be completed by spring 2010.

Please note that the IDS TCW activities are open to participation and anyone interested is welcome to join. Please contact any of us in case you want to be involved. Further details about these studies can be found on the IDS-TCW website.

Nanette Schloot and Roberto Mallone  
on behalf of The IDS TCW Committee