FOCiS®
Federation of Clinical Immunology Societies
www.focisnet.org

# FOCiS® 2012

Vancouver, BC, Canada ▪ June 20 - 23

www.focisnet.org

Abstract Supplement

# FOCIS 2012
# Abstract Supplement: Abstracts by Category

Federation of Clinical Immunology Societies
12th Annual Meeting
Vancouver, BC, Canada
June 20 - 23, 2012

## Table of Contents

differentiation. Using a blocking anti-TIM-3 mAb accelerated allograft rejection in the presence of host $CD4^+$ T cells. Here we examined the expression and function of TIM-3 on human regulatory T cells and its role in autoimmune diseases. Our results show that TIM-3 is not expressed on human Tregs *ex vivo*, but is upregulated both at an mRNA and protein expression levels soon after activation. Moreover, blocking TIM-3 interaction with its ligand Galectin-9 using an anti-TIM-3 mAb modulates the suppression capacity of Tregs. Finally, our preliminary results show that TIM-3 blockade during the course of activation of Tregs increases the level of expression of IL-17, suggesting a role of TIM-3 on the reprogramming of Tregs into Th17-Tregs. Thus, our results suggest that TIM-3 expression on Tregs plays a role in the regulation of immune tolerance and homeostasis.

**T.73. Determination of the Generation and Functionalities of Coxsackievirus B3-Specific T Cells Using MHC Class II Tetramers in the Murine Model of Viral Myocarditis**

*Arunakumar Gangaplara, Chandirasegaran Massilamany, Asit Pattnaik, David Steffen, Jay Reddy. University of Nebraska-Lincoln, Lincoln, NE*

Enteroviruses like Coxsackievirus B3 (CVB) are common suspects in patients with myocarditis/dilated cardiomyopathy (DCM). CVB can induce myocarditis in A/J mice, but the infectious virus becomes undetectable 14-18 days post inoculation. A long-standing question exists as to whether the chronic inflammatory process occurs due to an autoimmune response to cardiac antigens or to a viral-specific immune response. To dissect this complexity, we recently created major histocompatibility complex class II/$IA^k$ tetramers for cardiac myosin heavy chain-α (Myhc) 334-352 to determine the role of Myhc-reactive CD4 T cells in the development of viral myocarditis induced with CVB. We now report the creation of similar reagents for CVB. Thirty overlapping peptides of 20-mers spanning the entire region of viral protein (VP) 1 were designed and tested for their ability to stimulate splenocytes harvested from CVB-infected mice, leading us to identify three immunogenic peptides: VP1 681-700, VP1 721-740, and VP1 771-790. We then created $IA^k$ tetramers for these peptides and validated them using lymph node cell cultures prepared from A/J mice immunized with the corresponding peptides. Using splenocytes obtained from CVB-infected mice, we noted that, while all the three peptides induced proliferative responses, only VP1 721-740 and VP1 771-790 were found to bind antigen-sensitized CD4 T cells with specificity suggesting the generation of virus-reactive T cells with variable affinities. In conjunction with Myhc 334-352 tetramers, the availability of CVB tetramers permit the generation and functionalities of self (Myhc)- and foreign (CVB)-reactive CD4 T cells in the causation of post infectious myocarditis induced by CVB.

**T.74. Anti-inflammatory Activity of Artemisia Absinthium L. in Viper Venom (Montivipera Xanthina (Gray) and Carreganen-Induced Acute Inflammation in Rat**

*Ayse Nalbantsoy[1], Sura Baykan Erel[1], Cinel Koksal[1], Bayram Gocmen[1], Mehmet Zulfu Yildiz[2], Ulku Karabay Yavasoglu[1]. [1]Ege University, Izmir, Turkey; [2]Harran University, Sanlıurfa, Turkey*

Artemisia is represented by 23 species in the Turkish flora, and 6 of them are naturally distributed in western and southwestern Anatolia. Artemisia absinthium L. has been used worldwide in folk medicine since ancient times. The present study was conducted to explore the anti-inflammatory effects of A. absinthium L. methanolic extract in the rat hind paw model of Montivipera xanthina (Gray) venom and carrageenan-induced acute inflammations. Wistar rats were treated intraperitoneally with saline (as control group), 10 mg/kg indomethacin (as the positive control drug) and A. absinthium L. extract (12.5 or 25, 50 and 100 mg/kg), 30 min before venom or carrageenan was injected in saline

subplantarly into the left hind paw. Paw volume was measured by water plethysmometer before and 0.5, 1, 2, 3 and 4 h after the injection of venom or carrageenan. As a result, intraperitoneal administration of 50 mg/kg extract inhibited venom induced paw swelling at 0.5, 1, 2 and 3 h ($p<0.05$) while 12.5, 25 and 50 mg/kg extract inhibited carrageenan-induced paw swelling at 2, 3, 4 and 5 h ($p<0.05$). A. absinthium extract showed significantly better activity at doses of 50 mg/kg than indomethacine (10 mg/kg) ($p<0.05$) in both inflammation models. In conclusion, the methanolic extract of A.absinthium contains several components able to inhibit or inactivate toxins in Montivipera xanthina venom for the edema formation. This is the first report on Montivipera xanthina venom and A. absinthium extract as an inflammatory and anti-inflammatory agent respectiviely.

**T.75. Activated Memory T Cells: Biological Marker for Clinical Disease Progression in SLE**

*Chaim Jacob. University of Southern California, Los Angeles, CA*

We found that the $CD4^+$ $CD44^{high}$ $CD62L_{low/neg}$ subset of T cells correlate better than most other biological surrogate markers with the development of clinical disease in SLE mice. As determined by gene-array analysis, we have shown that these activated memory T cells display neither a Th1 nor Th2 profile; rather they express a Th17 profile and produce IL-17. These cells are refractory to anti-CD3-induced apoptosis *in vitro* and express apoptosis inhibiting genes. Most importantly, these activated memory cells progressively increase in numbers during disease development, not only in the spleen, but in the kidney as well. Perfusion of mouse with Dynabeads causes the beads to become stuck in the kidney glomeruli which enables the collection of pure glomeruli by a magnetic concentrator and reliably separate the glomeruli from the tubulo-interstitial compartment of the kidney. Using a complex set of enzymatic, and mechanical separations, we can isolate from both glomeruli and tubulo-interstitial compartment (each separately), living lymphocytes and macrophages, and submit these live cell to quantitative flow cytometric analysis. Using this method we demonstrate that, with disease progression, the activated memory T cells represent the majority of $CD4^+$ T cells in the glomeruli and the tubulo-interstitial compartment of kidneys of lupus mice.

**T.76. The Nature of Modified LDL Involved in the Formation of Immune Complexes has a Modulating Effect on Macrophage Activation**

*Gabriel Virella[1], Yanchun Li[1], Yan Huang[1], Maria Lopes-Virella[1,2]. [1]Medical University of South Carolina, Charleston, SC; [2]Ralph A. Johnson VA Medical Center, Charleston, SC*

The involvement of modified LDL-immune complexes (IC) in CAD progression is supported by a variety of data. Studies in type 1 diabetes showed that high levels of IC containing predominantly oxidized LDL (oxLDL) were strongly associated with CAD progression. In contrast, in type 2 diabetes, high levels of IC containing predominantly malondialdehyde (MDA)-LDL were significantly associated with the occurrence of acute cardiovascular events, especially MI. We have carried out studies comparing the effects of oxLDL-IC and MDA-LDL-IC in human macrophages to determine whether the nature of the modified LDL modulated the effects of FcγRI engagement by LDL-IC. Human macrophages were incubated with oxLDL and MDA-LDL and also with oxLDL-IC and MDA-LDL-IC. Free oxLDL and MDA-LDL did not change IL-6, MMP-9 and TIMP-1 mRNA expression but led to a significant increase in MCP-1 mRNA expression. OxLDL-IC and MDA-LDL-IC induced similar high-level mRNA expression of MCP-1 and MMP-9 but the expression of IL-6 was considerably higher after incubation with oxLDL-IC. No effect was observed on TIMP-1 mRNA. In the same experiments, levels of IL-6, MCP-1 and TIMP-1 released into the culture medium mimic those obtained by PCR. Interestingly the levels of MMP-9 released by macrophages exposed to MDA-