BROAD REPERTOIRE OF T CELL AUTOREACTIVITY DIRECTLY FROM ISLETS OF DONORS WITH TYPE 1 DIABETES (T1D)

Jenny Aurielle B. Babon[1], Megan E. DeNicola[1], David M. Blodgett[1], Inne Crèvecoeur[2], Thomas S. Buttrick[3], René Maehr[4], Rita Bottino[5,6], Ali Naji[7], John Kaddis[8], Wassim Elyaman[3], Eddie A. James[9], Rachana Haliyur[10], Marcela Brissova[10], Lut Overbergh[2], Chantal Mathieu[2], Thomas Delong[11], Kathryn Haskins[11], Alberto Pugliese[12], Martha Campbell-Thompson[13], Clayton Mathews[13], Mark A. Atkinson[13], Alvin C. Powers[10,14,15], David M. Harlan[1], Sally C. Kent[1]

[1]Division of Diabetes, Diabetes Center of Excellence, Department of Medicine, University of Massachusetts Medical School; [2]Laboratory for Clinical and Experimental Endocrinology, Department of Clinical and Experimental Medicine, KU Leuven, Leuven, Belgium; [3]Ann Romney Center for Neurologic Diseases, Brigham and Women's Hospital and Harvard Medical School, Boston, MA; [4]Program in Molecular Medicine, Diabetes Center of Excellence, University of Massachusetts Medical School; [5]Institute of Cellular Therapeutics, Allegheny-Singer Research Institute, Pittsburgh, PA; [6]Department of Biological Sciences, Carnegie Mellon University, Pittsburgh, PA; [7]Institute for Diabetes, Obesity, and Metabolism, University of Pennsylvania School of Medicine, Philadelphia, PA; [8]Department of Information Sciences, Beckman Research Institute, City of Hope, Duarte, CA; [9]Benaroya Research Institute at Virginia Mason, Seattle, WA; [10]Division of Diabetes, Endocrinology and Metabolism, Department of Medicine, Vanderbilt University Medical Center, Nashville, TN; [11]Department of Immunology and Microbiology, University of Colorado School of Medicine, Denver, Anschutz Medical Campus, Aurora, CO; [12]Diabetes Research Institute, University of Miami, Miami, FL; [13]Departments of Pathology, Immunology, and Laboratory Medicine, University of Florida, Gainesville, FL; [14]Department of Molecular Physiology and Biophysics, Vanderbilt University, Nashville, TN; [15]VA Tennessee Valley Healthcare System, Nashville, TN

Type 1 diabetes (T1D) is an autoimmune disease characterized by the infiltration of lymphocytes into the insulin-producing β-cells in the pancreas. We have isolated live T cells sorted or grown directly from the isolated, handpicked islets of human donors with T1D. We received ~500 islet equivalent EQ of variable purity (10-90%) from 12 donors with T1D (disease duration 0.42-20 years) and from seven control donors and two donors with type 2 diabetes (T2D). A total of 321 T cell lines and clones were derived from the islets of donors with T1D (3 lines from the 9 control donors). These are 131 CD4+ lines and clones, 47 CD8+ lines and 143 lines that contain both CD4+ and CD8+ T cells. From 50 lines and clones examined to date, we have determined the autoreactivity of 19 and have seen a broad repertoire of T cell autoreactivity in the islets, including characterized targets and post-translationally modified targets. Autoreactivity of CD4+ T cell lines was to three different peptides from glutamic acid decarboxylase 65 (GAD; $GAD_{115-127}$, $GAD_{274-286}$, $GAD_{555-567}$), $proinsulin_{76-90}$, and to chromogranin A or proinsulin expressed by DR4+DQ8+ B cells transduced with lentivirus containing constructs with the open reading frames corresponding to whole autoantigens. Reactivity to modified peptides included the glucose-regulated protein 78 and islet amyloid polypeptide with arginine to citrulline modifications ($GRP78_{292-305(Arg\text{-}Cit297)}$ and $IAPP_{65-84(Arg\text{-}Cit\ 73,\ 81)}$), deaminations ($IA\text{-}2_{545-562(Gln\text{-}Glu\ 548,\ 551,\ 556)}$, and to several insulin hybrid peptides. These autoreactive CD4+ T cell lines and clones secreted only pro-inflammatory cytokines (IFN-γ, TNFα) upon peptide stimulation. For CD8+ T cells from islets, from one donor with T1D, we saw binding of a pool of HLA-A2 pentamers loaded with insulin B_{10-18}, $IA\text{-}2_{797-805}$ and insulin specific glucose-6-phosphatase catalytic subunit related protein, $IGRP_{265-273}$. These results have implications for the development of successful prevention and reversal therapeutic strategies in T1D.

Contact:
Jenny Aurielle B. Babon, Ph.D.
University of Massachusetts Medical School
Jenny.babon@umassmed.edu