Finally, the temporal dimension of hepatocyte regeneration was queried. Using APAP-ALF mice, the researchers found a temporal disconnect between wound healing and hepatocyte proliferation, with the latter peaking more than 24 hours after most of the wound had healed, supporting the hypothesis that hepatocytes responsible for wound healing do not arise because of hepatocyte proliferation. Four-dimensional intravital microscopy subsequently demonstrated migration of the hepatocyte sheet, with the previously described migratory hepatocytes appearing at the leading edge. These findings were validated with knockdown of ANXA2 in hepatocytes, which led to reduced wound closure.

The key findings from this work suggests that a novel migratory ANXA2$^+$ hepatocyte population emerges after acute liver injury, at the edge of necrosis, to enable collective migration of the hepatocyte sheet and heal the necrotic region; a process that is dependent on ANXA2 expression. Although the authors reported finding ANXA2$^+$ hepatocytes in an array of acute and chronic liver diseases, their role in hepatocyte regeneration in ALF that recovers and does not require liver transplantation, or in normal liver regeneration independent from liver injury remains to be elucidated. In addition, their role in liver regeneration seen with chronic liver diseases is yet to be determined. Although it remains to be seen whether these newly identified migratory hepatocytes can be manipulated for clinical benefit in our patients, these findings challenge previously accepted concepts of liver regeneration and represent a significant shift in our understanding of hepatocyte physiology.

HARRY SUTTON
Division of Gastroenterology, Hepatology, and Nutrition
Hospital for Sick Children and University of Toronto
Toronto, Ontario, Canada

MAMATHA BHAT
Ajmera Transplant Centre *and*
Division of Gastroenterology and Hepatology
University Health Network *and*
Department of Medicine
University of Toronto
Toronto, Ontario, Canada

Conflicts of interest
The authors disclose no conflicts.

Defining the Role of Immune Microniches in Intestinal Effector Regulatory T-Cell Functionality

Gu Y, Bartolomé-Casado R, Xu C, et al. Immune microniches shape intestinal T_{reg} function. Nature 2024;628:854–862.

Regulatory T cells (Tregs) in the intestinal milieu orchestrate tolerance to commensal microbiota and self-antigens, while also contributing to host defense against invading pathogens. Imbalances of Treg function can lead to compromised immunity and intestinal inflammation. Whereas the secondary lymphoid organs are known to be crucial for Treg cell induction, the primary anatomic sites, cellular neighborhoods, and transcriptional programs responsible for the induction and maintenance of effector Treg (eTreg) suppressor functions remain poorly understood.

Gu et al provide critical insights into the immune niches and spatial compartmentalization that shape microorganism-reactive programming of eTreg cell function. To explore the temporal and spatial development of eTreg function in the intestinal microenvironment in the context of tolerance and inflammation, the authors used mice infected with *Helicobacter hepaticus* (*Hh*), a pathobiont that establishes long-term colonization in the large intestine, and then adoptively transferred naïve T cells with transgenic receptors (TCRs) that recognize *Hh* in these mice. In contrast to the existing paradigm, the authors demonstrated that the lamina propria, rather than secondary lymphoid tissues, serves as the critical microniche for supporting eTreg cell functions, as evidenced by enhanced T-cell proliferation, Treg cell differentiation, TCR signaling, and interleukin-10 (IL-10) production. Using 2-photon photo-activation labeling and single-cell RNA sequencing (NICHE-seq), the authors demonstrated that lymphoid-associated central Treg cells and tissue-resident eTreg cell populations are transcriptionally and spatially distinct. Using antibodies to block IL-10 receptor–mediated signaling to promote inflammation in the model, these studies suggested that Treg cell phenotypes in the lamina propria are stable upon establishment of the tolerogenic niche and that additional suppressor functions of eTreg cells appear to be constrained by local responses independently from IL-10. Detailed mapping of the microniches through 2-photon live imaging, single-cell profiling, and immunofluorescence identified serial interactions of motile IL-10–producing eTreg cells with CD206$^+$ macrophages and identified receptor-ligand pairs that may govern the putative tolerogenic interaction between CD206$^+$ macrophages and eTreg cells in the lamina propria. In the context of inflammation, several cell types, in particular CD103$^+$SIRPα^+ dendritic cells, that were confined to the lymphoid-associated tissue during homeostasis, were recruited to the lamina propria during inflammation, suggesting that dendritic cells may disrupt the homeostatic interactions within the lamina propria.

This sophisticated study advances our understanding of how immune environments within the gut shape the microorganism-reactive function, motility, and stability of eTreg cells. The study not only expands our understanding of the cellular mechanisms underlying immune tolerance of Tregs in the gut, but also offers a promising framework for developing targeted therapies that harness the properties of immune microniches to enhance Treg function and promote intestinal health. Future studies are required to elucidate the underlying mechanisms and interactions in lamina

propria microniches, to evaluate whether the findings are generalizable to other TCR specificities or pathobiont models, and to explore the therapeutic potential of manipulating these immune niches in human health and disease.

DANIEL KOTLARZ
Department of Pediatrics
Dr von Hauner Children's Hospital
University Hospital
Ludwig-Maximilians-Universität Munich
Munich, Germany, *and*
Institute of Translational Genomics
Helmholtz Zentrum München
German Research Center for Environmental Health
Neuherberg, Germany

Conflicts of interest
The author discloses no conflicts.