LIVER DIGEST

February 17, 2023

A bi-weekly update of PLRC happenings

PITTSBURGH LIVER RESEARCH CENTER
A partnership of University of Pittsburgh & UPMC

In this issue:

Faculty Highlight

Haitao Guo, PhD., Professor, Department of Microbiology and Molecular Genetics, and colleagues published an article in Antiviral Research entitled "***Screening of an epigenetic compound library identifies BRD4 as a potential antiviral target for hepatitis B virus covalently closed circular DNA transcription.***"

https://doi.org/10.1016/j.antiviral.2023.105552

HBV cccDNA is the persistent form of viral genome, which exists in host cell nucleus as an episomal minichromosome decorated with histone and non-histone proteins. cccDNA is the authentic viral transcript template and resistant to current antivirals. Growing evidence shows that the transcriptional activity of cccDNA minichromosome undergoes epigenetic regulations, suggesting a new perspective for anti-cccDNA drug development through targeting histone modifications. In this study, we screened an epigenetic compound library in the cccDNA reporter cell line HepBHAe82, which produces the HA-tagged HBeAg in a cccDNA-dependent manner. Among the obtained hits, a bromodomain-containing protein 4 (BRD4) inhibitor MS436 exhibited marked inhibition of cccDNA transcription in both HBV stable cell line HepAD38 and HepG2-NTCP or primary human hepatocyte infection system under noncytotoxic concentrations. Chromatin immunoprecipitation (ChIP) assay demonstrated that MS436 dramatically reduced the enrichment of H3K27ac, an activating histone modification pattern, on cccDNA minichromosome. RNAseq differential analysis showed that MS436 does not drastically change host transcriptome or induce any known anti-HBV factors/pathways, indicating a direct antiviral effect of MS436 on cccDNA minichromosome. Interestingly, the MS436-mediated inhibition of cccDNA transcription is accompanied by cccDNA destabilization in HBV infection and a recombinant cccDNA system, indicating that BRD4 activity may also play a role in cccDNA maintenance. Furthermore, depletion of BRD4 by siRNA knockdown or PROTAC degrader resulted in cccDNA inhibition in HBV-infected HepG2-NTCP cells, further validating BRD4 as an antiviral target. Taken together, our study has demonstrated the practicality of HepBHAe82-based anti-HBV drug screening system and provided a proof-of-concept for targeting HBV cccDNA with epigenetic compounds.

Please acknowledge all support from the PLRC in your publications and presentations. (NIH/NIDDK P30DK120531)

Please continue to share your relevant accolades (grants, publications, awards and other news worthy items) with us, as it relates to the PLRC mission, so we can share with all of our members.

Please visit the PLRC website (www.livercenter.pitt.edu) for up-to-date news, and upcoming seminar and event information. Contact Aaron Bell (bellaaro@pitt.edu) if you have specific questions or suggestions.

Our mailing address is:

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PLRC SEMINARS

- **February 21 12:00 pm – 1:00 pm *HYBRID***
  S120 BST & via Zoom

Hossam A. Abdelsamed, PhD
Research Assistant Professor
Department of Surgery, Starzl Transplant Institute

Title: Human T cell Biology: From Epigenetics to Cross-talk and Liver Autoimmunity

- **February 28 12:00 pm – 1:00 pm *HYBRID***
  S120 BST & via Zoom

Hun-Way Hwang, MD, PhD
Assistant Professor
Department of Pathology

Title: Alternative polyadenylation: regulation of gene function and isoform expression

- **March 7 @ 12:00 pm – 1:00 pm *HYBRID***
  S120 BST & via Zoom

George Michalopoulos, MD, PhD
Professor and Chair
Department of Pathology

TITLE: TBD

Faculty Highlight

Zachary Freyberg, MD, PhD, Associate Professor in the Departments of Psychiatry and Cell Biology and former PLRC P&F awardee along with Silvia Liu, Evan Delgado, Aaron Bell and Paul Monga published an article in Am. J. Pathology, entitled, "A spatial atlas of Wnt receptors in adult mouse liver".

Abstract:

Hepatic zonation is critical for most metabolic functions in liver. Wnt signaling plays an important role in establishing and maintaining liver zonation. Yet, the anatomic expression of Wnt signaling components, especially all 10 Frizzled receptors (Fzds), has not been characterized in adult liver. To address this, we quantitatively mapped the spatial expression of Fzd receptors in adult mouse liver via multiplex fluorescent in situ hybridization. While all 10 Fzds are expressed within a metabolic unit, Fzds 1, 4, and 6 are the highest expressed. Though the majority of Wnt signaling occurs in Zone 3, expression of most Fzds is not zonated. In contrast, Fzd6 is preferentially expressed in Zone 1. We also verified that Wnt2 and Wnt9b expression is highly zonated and primarily found in Zone 3. Therefore, our results suggest that zonated Wnt/β-catenin signaling at baseline is mostly due to Wnt2 and Wnt9b rather than zonation of Fzd mRNA expression. Finally, we showed that Fzds and Wnts are not uniformly expressed by all hepatic cell types. Instead, there is broad distribution among both hepatocytes and non-parenchymal cells, including endothelial cells. Overall, our establishment of a definitive mRNA expression atlas, especially of Fzds, opens the door to future functional characterization in healthy and disease liver states.

Gayden J, Hu S, Joseph PN, Delgado E, Liu S, Bell A, Puig S, Monga SP, Freyberg Z. *A spatial atlas of Wnt receptors in adult mouse liver.* Am J Pathol. 2023 Feb 9:S0002-9440(23)00044-5. PMID: **36773785** DOI: 10.1016/j.ajpath.2023.01.011

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Faculty Highlight

Dr. Michael Oertel, PhD Dr. Mo Ebrahimkhani, MD.

Drs. Michael Oertel and Mo Ebrahimkhani co-authored a review article in Cells entitled, "*Therapeutic Cell Repopulation of the Liver: From Fetal Rat Cells to Synthetic Human Tissues*".

Shafritz DA, Ebrahimkhani MR, Oertel M. Therapeutic Cell Repopulation of the Liver: From Fetal Rat Cells to Synthetic Human Tissues. Cells. 2023; 12(4):529. https://doi.org/10.3390/cells12040529

Abstract

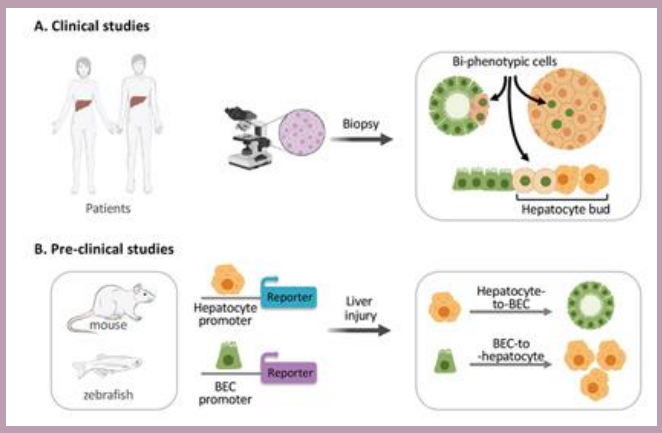
Progenitor cells isolated from the fetal liver can provide a unique cell source to generate new healthy tissue mass. Almost 20 years ago, it was demonstrated that rat fetal liver cells repopulate the normal host liver environment via a mechanism akin to cell competition. Activin A, which is produced by hepatocytes, was identified as an important player during cell competition. Because of reduced activin receptor expression, highly proliferative fetal liver stem/progenitor cells are resistant to activin A and therefore exhibit a growth advantage compared to hepatocytes. As a result, transplanted fetal liver cells are capable of repopulating normal livers. Important for cell-based therapies, hepatic stem/progenitor cells containing repopulation potential can be separated from fetal hematopoietic cells using the cell surface marker δ-like 1 (Dlk-1). In livers with advanced fibrosis, fetal epithelial stem/progenitor cells differentiate into functional hepatic cells and out-compete injured endogenous hepatocytes, which cause anti-fibrotic effects. Although fetal liver cells efficiently repopulate the liver, they will likely not be used for human cell transplantation. Thus, utilizing the underlying mechanism of repopulation and developed methods to produce similar growth-advantaged cells in vitro, e.g., human induced pluripotent stem cells (iPSCs), this approach has great potential for developing novel cell-based therapies in patients with liver disease. The present review gives a brief overview of the classic cell transplantation models and various cell sources studied as donor cell candidates. The advantages of fetal liver-derived stem/progenitor cells are discussed, as well as the mechanism of liver repopulation. Moreover, this article reviews the potential of in vitro developed synthetic human fetal livers from iPSCs and their therapeutic benefits.

Faculty Highlight

Donghun Shin, PhD., Associate Professor in the department of Developmental Biology has done fundamental work in understanding liver development and cellular differentiation. He recently published an article in Hepatology entitled, "*Hepatocyte-to-cholangiocyte conversion occurs through transdifferentiation independently of proliferation in zebrafish.*" In addition he co-authored a review in Seminars in Liver Disease entitled, "*Update on Hepatobiliary Plasticity*".

Lee SH, So J, Shin D. Hepatocyte-to-cholangiocyte conversion occurs through transdifferentiation independently of proliferation in zebrafish. Hepatology. 2023 Jan 3. doi: 10.1097/HEP.0000000000000016. Epub ahead of print. PMID: 36626626.

Kim M, Rizvi F, Shin D, Gouon-Evans V. Update on Hepatobiliary Plasticity. Semin Liver Dis. 2023 Feb 10. doi: 10.1055/s-0042-1760306. Epub ahead of print. PMID: 36764306.https://www.thieme-connect.com/products/ejournals/html/10.1055/s-0042-1760306



The liver field has been debating for decades the contribution of the plasticity of the two epithelial compartments in the liver, hepatocytes and biliary epithelial cells (BECs), to derive each other as a repair mechanism. The hepatobiliary plasticity has been first observed in diseased human livers by the presence of biphenotypic cells expressing hepatocyte and BEC markers within bile ducts and regenerative nodules or budding from strings of proliferative BECs in septa. These observations are not surprising as hepatocytes and BECs derive from a common fetal progenitor, the hepatoblast, and, as such, they are expected to compensate for each other's loss in adults. To investigate the cell origin of regenerated cell compartments and associated molecular mechanisms, numerous murine and zebrafish models with ability to trace cell fates have been extensively developed. This short review summarizes the clinical and preclinical studies illustrating the hepatobiliary plasticity and its potential therapeutic application.

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A train derailment on Friday February 3rd, in East Palestine Ohio, 50 miles from Pittsburgh, PA, resulted in a chemical spill from tanker cars, massive fire and evacuation of nearby residents. PLRC Faculty Juliane Beier, PhD who studies vinyl chloride and its effects on liver disease, is considered and expert in the field and was interviewed by NPR radio , CBS News and WTAE television. See links to those interviews below. Dr. Beier received prior funding from the Pittsburgh Liver Research Center Pilot & Feasibility program to study the effects of vinyl chloride exposure on liver cancer, the results of which lead to further funding by the National Institutes of Health.

Read more on how Vinyl Chloride, the chemical used to make PVC plastics, which was released from the Ohio train derailment, can damage the liver and exacerbate underlying disease in "The Conversation".

NPR interview: **NEWS story on WTAE:**
CBS News : **The Conversation:**

ANNOUNCEMENTS

CLA FEBRUARY EVENTS: (https://communityliveralliance.org/)

2/24/23 CLA/WVU Liver Update Treating HCV in primary Care setting

Funding Opportunities

- **Gilead Research Scholars-Liver Disease program. Junior Faculty . May 1, 2023**
- **To see all NIH Grants sorted by week, please visit: NIH Guide: 2022** Or click below:

Week of: **Feb 3** **Feb 10** **Feb 17**

Click here for all current NIDDK Funding opportunities

WANT ADS

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  Please send any "wants/needs" to be advertised to bellaaro@pitt.edu (Aaron Bell)
- Postdoc/Technician: Bone cell Differentiation $45K; PI: Harry Blair: US citizen ONLY!
  Contact: 412-383-9616 or hcblair@pitt.edu