Annual Joseph E. and Martha E. Kutscher Digestive Disease Research Research Symposium

October 8 & 9, 2015
Texas A&M Health Science Center College of Medicine
Medical Education Center LH2
Temple, Texas
Welcome

Welcome to the annual Joseph E. and Martha E. Kutscher Baylor Scott & White Digestive Disease Research Center (DDRC) symposium and the 4th DDRC symposium. We are excited to have two keynote speakers: Dr. Satdarshan Monga who will deliver our basic science keynote lecture and Dr. David Cohen that will present at Internal Medicine Grand Rounds. Our schedule also includes numerous other leaders in the field of gastrointestinal disorders. We are also pleased to be able to showcase some of the exciting research currently underway in the S&W DDRC and in the laboratories of our collaborators and co-workers. We hope this symposium will serve as a springboard to foster additional successful collaborations.

Gianfranco Alpini, Ph.D. Richard Erickson, M.D.
Basic Science Director Clinical Director

Acknowledgements:

The DDRC staff and faculty wish to acknowledge the following people for their assistance in the planning and development of this symposium:

Dr. Kathryn J. Kotrla (Joseph E. & Martha E. Kutscher’s niece)
Mrs. Melody Ivy
Mr. Glen Cryer
Mr. Gary Hansen
Pictured from left to right: (bottom row) Gianfranco Alpini, Ph.D., Shannon Glaser, Ph.D., Dawn Sears, M.D., (top row) Sharon DeMorrow, Ph.D., Heather Francis, Ph.D. Fanyin Meng, M.D., Ph.D., Richard Erickson, M.D., and Haibo Bai, Ph.D., (not shown: Terry Lairmore, M.D., Walter P. Dyck, M.D. & Lucas Wong, M.D.).

*About the Baylor Scott & White DDRC*

Our major goal and objective is to discover and share cutting edge knowledge about the digestive system and the diseases that affect millions of lives in the United States. Our research team consists of both basic scientists and physician scientists from a wide variety of backgrounds with varying interests and disciplines. These combined investigators support the practice of “bench to bedside” research and translational medicine to benefit and contribute to human health. Along with seasoned investigators, the Baylor Scott & White DDRC also believes in the training of future scientists and provides an excellent learning environment for the training of undergraduates, medical students, graduate students, postdoctoral fellows, residents and clinical fellows. Our basic and clinical faculty is currently funded by the NIH and the VA along with funding from other private foundations including: PSC Partners Seeking a Cure and the Ladies Auxiliary of Texas for VFW.
Mission statement

The mission of the Scott & White DDRC is to enhance our understanding of digestive system diseases through the integration of basic and clinical science research to provide new strategies to diagnose, treat and cure diseases of the digestive tract.
The Nicholas C. Hightower Endowed Chair of Gastroenterology

In 1952 Dr. Nicholas C. Hightower was recruited to Scott & White Hospital from the Mayo Clinic in Rochester. Dr. Hightower was a member of the Division of Gastroenterology and Professor of Medicine within Texas A&M. He served as the Chairman for Clinical Pathology at Scott & White. Dr. Hightower created both the first GI fellowship program and the first GI Research Program at Scott & White. He served as the director of the Division of Research and Education from 1968-1978 and was an active investigator himself. During his career he was also the first investigator to receive extramural funding at Scott & White and continued to be funded throughout his career. In 2002, Dr. Gianfranco Alpini, Basic Science Director of the DDRC was named as the holder of the Endowed Centennial Chair in Gastroenterology named in honor of Dr. Hightower. Recruited in 1994, Dr. Alpini began his research on the VA campus and, over the years, has gained the reputation for being a leader in the field of cholangiocyte biology. His dedication, coupled with the support of the Hightower chair, has enabled Dr. Alpini and the DDRC faculty to produce consistent, high quality research while building a well-recognized GI research program. Thus carrying on the tradition of research excellence started by Nicholas Hightower and supported by the Kutscher family.

Dr. Hightower at his retirement reception (above) and restoring an automobile (below).
Dr. Hightower outside of the first research building

Dr. Hightower and colleagues in the GI Research Lab, Desk G, Scott & White
Joseph E. and Martha E. Kutscher

Martha Kutscher enjoying a sunset at the “Ranch”

Joseph E. and Martha E. Kutscher were longtime patients of Dr. Hightower’s and, over the years, Joe and Martha became close friends with “Doc” and his wife, DonAnne. The Kutschers and the Hightowers enjoyed many fishing trips together in Wyoming and developed a lifelong friendship that is now honored by the Nicholas C. Hightower Centennial Chair.

Joe and Martha married shortly after Joe served in World War II. They were partners in developing Continental Homes, a real estate development company that spread throughout Southeast and Midwest America. Martha, having been born and raised in Taylor, Texas, turned Joe into a transplanted Texan. In the early 1950’s, they acquired over 4,000 acres in what was then isolated Texas Hill Country northwest of Austin. Now the “Ranch”, as the family calls Kutscher Ranch, is minutes from Austin, but still offers an unparalleled natural respite, with its hills, creeks, springs, and native American artifacts. Joe used the “Ranch” as a personal hunting reserve for his friends, including Darrell Royall and Doc. Martha passed away October 22, 2012 and will always be remembered through her generosity and legacy.

In addition to her loyal support of Scott & White Healthcare, she and her niece, Dr. Kathryn Kotrla, donated part of the “Ranch” to fund one of the buildings on the new Austin Children’s Shelter campus.
IM Grand Rounds Keynote Speaker:

Dr. David Cohen, M.D., Ph.D.

For almost two decades, Dr. Cohen’s research program has focused on understanding the molecular regulation of hepatic lipid and glucose metabolism. Among his contributions has been to describe novel roles for phosphatidylcholine transfer protein (PC-TP) in the control of hepatic lipid and glucose homeostasis. Because it binds phosphatidylcholines with high specificity and catalyzes their transfer between membranes in vitro, he originally proposed that PC-TP might play a role in the hepatocellular trafficking of biliary phospholipids to the canalicular membrane for secretion into bile and to the sinusoidal plasma membrane for incorporation into high density lipoprotein (HDL) particles. In the course of these studies, he was the first to clone PC-TP, to characterize the gene and its transcription, and to express recombinant protein that we used to elucidate membrane-binding domains. The Cohen laboratory crystallized PC-TP in complex with phosphatidylcholine, leading to our report of the three dimensional structure. Studies in cell culture systems and Pctp−/− mice revealed much broader functions than anticipated in regulating metabolism. In collaborative studies, Dr. Cohen utilized hyperinsulinemic euglycemic clamp studies to demonstrate that Pctp−/− mice are highly sensitized to insulin action and are protected against diet-induced diabetes due to suppression of hepatic glucose production. His laboratory further showed that Pctp−/− mice exhibit increased adaptive thermogenesis due to increased sensitivity of brown adipocytes to stimulation by norepinephrine. The Cohen Laboratory discovered small molecule inhibitors of PC-TP, which show promise for increasing insulin sensitivity in cells and in mice. In separate studies, the laboratory demonstrated that PC-TP binds and activates thioesterase superfamily member (Them) 2, a newly described fatty acyl-CoA thioesterase. We have also demonstrated key roles of both Them1 and Them2 in regulating hepatic lipid and glucose metabolism, as well as energy homeostasis.

Dr. Cohen is the Director of Hepatology at Brigham and Women’s Hospital, Director of Harvard-MIT Division of Health Sciences and Technology and holds the Robert H. Ebert Professor of Medicine and Health Sciences and Technology, Harvard Medical School.
Basic Science Keynote Speaker:

Dr. Satdarshan (Paul) S. Monga, M.D.

Dr. Monga’s laboratory is focused on understanding the molecular mechanisms of liver growth and development in health and disease especially trying to address the molecular basis of liver development, growth, regeneration and cancer. Several signaling pathways have been identified to direct such events including the Wnt/β-catenin, HGF/Met, PDGFRβ and others.

Liver development in mice is initiated at around E8-8.5 stages of gestational development. Once foregut endoderm gains ‘competence’, hepatic signatures are initiated during the process of ‘induction’. The primitive liver bud contains bipotential stem cells or progenitors, which undergo expansion and regulated differentiation into hepatocytes and biliary epithelial cells during the process of ‘morphogenesis’. One of the major focuses of Monga laboratory is to identify the molecular basis of hepatic morphogenesis. More specifically how does the hepatic progenitor or the bipotential stem cell undergo self-renewal (symmetric division), lineage specification and differentiate further towards primitive bile duct cells or immature hepatocytes (asymmetric division) and then to fully differentiated cells. Using conditional null mice, embryonic liver cultures and other modalities, the lab is investigating the roles, regulation and interactions of various pathways, which will not only further our understanding of this fundamental process of biology, but might also provide insight into the molecular basis of disease that recapitulates development in adulthood-hepatocellular cancer (HCC).

HCC is the third leading cause of death due to cancers and remains a disease with poor treatment options. A significant focus in Dr. Monga’s laboratory is towards targeting this pathway and others, which are normally upregulated during liver development at the time of peak proliferation and stem cell renewal, as a novel therapeutic measure.

In addition, various animal models have been generated in Dr. Monga's laboratory, which conditionally overexpress or show lack of expression of important genes such as β-catenin and others, which are in the process of being studied for the role of canonical Wnt signaling in additional liver diseases such as alcoholic liver disease, nonalcoholic fatty liver disease, glucose metabolism and others.

Thus, the Monga lab is focused on understanding the molecular and cellular basis of normal liver characteristics such as development, regeneration, metabolism and growth as well as of liver pathologies such as neoplasms (HCC and hepatoblastoma), fibrosis, cirrhosis, alcoholic liver disease, non-alcoholic fatty liver disease and others. This incorporates studies on cell proliferation, adhesion, differentiation, invasion, apoptosis, metabolism and on stem cells in adult, fetal and embryonic livers.

Dr. Monga's research is and has been funded by the NIH (NIDDK, NCI, NHLBI), American Cancer Society and private pharmaceutical companies.
The Joseph E. and Martha E. Kutscher DDRC Annual Symposium
Thursday & Friday, October 8-9, 2015
MEC LH2/Mayborn Auditorium

Thursday, October 8th:
Poster session & wine/cheese reception
4:00 – 5:30 PM
MEC Lobby
Judges: Dr. Anatoliy Gashev, Dr. Cynthia Meininger and Dr. David Dostal
Awards will be given for 1st, 2nd, 3rd for both basic & clinical posters
4:00 – 5:30 poster presentations

5:30 – 6:00 PM
Introduction & Welcome
Dr. Gianfranco Alpini, Ph.D.
Mayborn Auditorium
Distinguished Professor of Medicine, Texas A&M Health Science Center
Nicholas C. Hightower Endowed Chair of Gastroenterology
Director, Digestive Diseases Research Center
Baylor Scott & White Healthcare
VA Career Scientist
Central Texas Veterans Health Care System

5:30 – 6:00 PM
Young Investigator podium presentations

6:00 – 7:00 PM
Satdarshan (Paul) S. Monga, M.D.
Keynote address
Vice Chair of Experimental Pathology
Endowed Chair for Experimental Pathology
Professor of Pathology and Medicine
Assistant Dean for Medical Scientist Training Program
University of Pittsburgh, School of Medicine
“Cell-Cell Junctions and Barriers: Function Beyond Cell Adhesion”

7:00 PM
Poster awards

Friday, October 9th:
Registration and reception
7:15 am – 7:45 am
Continental Breakfast
MEC LH2

7:45 am – 8:15 am
Opening remarks and introduction:
Dr. Kathryn Kotrla (Joseph & Martha Kutscher’s niece)
President, MEK
SESSION I: Molecular Mechanisms of Liver Repair

Moderators: Dr. Monga & Dr. Borad
MEC LH2
20 minutes presentation followed by 5 minutes discussion

8:15 – 8:40 am
Chandrashekhar Gandhi, Ph.D.
Professor, Medicine
Department of Pediatrics, Division of Gastroenterology, Hepatology and Nutrition
Cincinnati Children’s Hospital Medical Center
“Mitochondrial ALR deficiency and accelerated steatohepatitis”

8:40 – 9:05 am
Satdarshan (Paul) S. Monga, M.D.,
Vice Chair of Experimental Pathology
Endowed Chair for Experimental Pathology
Professor of Pathology and Medicine
Assistant Dean for Medical Scientist Training Program
University of Pittsburgh, School of Medicine
“Cell Molecule Wnt Signaling in Liver Regeneration”

9:05 – 9:30 am
Huiping Zhou, Ph.D.
Associate Professor
Microbiology & Immunology
Virginia Commonwealth University School of Medicine
“Conjugated bile acids promote cholangiocyte proliferation via activation of spingosine-1-phosphate receptor 2”

9:30 – 9:55 am  
Sharon DeMorrow, Ph.D.  
Associate Professor, Medicine  
Texas A&M Health Science Center  
Research Biologist, Central Texas Veterans Health Care System  
“Conjugated bile acids suppress the HPA axis via activation of hypothalamic glucocorticoid receptors in a rodent model of chronic biliary obstruction”

9:55 – 10:05 am  
Break

SESSION II: Carcinogenesis  
Moderators: Dr. Schwabe & Dr. Anant  
MEC LH2  
20 minutes presentation followed by 5 minutes discussion

10:05 – 10:30 am  
Shrikant Anant, Ph.D.  
Tom/Teresa Walsh Professor of Cancer Prevention Eminent Scholar  
Adjunct Professor, Department of Internal Medicine  
Adjunct Professor, Department of Surgery  
University of Kansas School of Medicine  
“Stem Cells and Colon Cancer: target for Prevention and Therapy”

10:30 – 10:55 am  
Fanyin Meng, M.D., Ph.D.  
Assistant Professor, Medicine  
Baylor Scott & White Healthcare, Texas A&M Health Science Center  
Research Biologist  
Central Texas Veterans Health Care System  
“Molecular Aspects of microRNAs in Gallbladder Stem Cells”

10:55 – 11:20 am  
Robert Schwabe, M.D.  
Associate Professor of Medicine  
Digestive and Liver Diseases  
Columbia University  
“Hepatocellular carcinoma originates from hepatocytes but not from liver progenitor cells”

11:20 – 11:45 am  
Mitesh Borad, M.D.  
Assistant Professor of Medicine  
Hematology/Oncology  
Mayo Clinic Cancer Center  
“Genomic Characterization of Cholangiocarcinoma”

11:45 am  
Please obtain your lunch from the MEC lobby and proceed to the Mayborn Auditorium.

12:00 pm  
Internal Medicine Grand Rounds
Mayborn Auditorium

David Cohen, M.D., Ph.D.
Director of Hepatology, Brigham & Women’s Hospital
Professor of Medicine & Health Sciences & Technology
Harvard Medical School
Director, Harvard-MIT Division of Health Sciences & Technology
“Pathogenesis and Management of Non-Alcoholic Fatty Liver Disease”

SESSION III: GI Disorders
Moderators: Dr. Dudeja & Dr. Glaser
MEC LH2
20 minutes presentation followed by 5 minutes discussion

1:10 – 1:35 pm
Pradeep Dudeja, Ph.D.
Professor of Physiology in Medicine
Director, Intestinal Transport Group
Gastroenterology & Hepatology
University of Illinois at Chicago
“All-trans-retinoic acid (ATRA): A novel potential therapeutic agent for inflammatory diarrhea"

1:35 – 2:00 pm
Didier Merlin, Ph.D.
Professor, Center for Diagnostics and Therapeutics, Center for Inflammation Immunity & Infection
Georgia State University
Research Career Scientist, VA Medical Center Decatur
“Edible Nanoparticles for IBD treatment?”

2:00 – 2:25 pm
Narendra Kumar, Ph.D.
Associate Professor
Pharmaceutical Sciences
Texas A&M Rangel College of Pharmacy
“Gut-feeling, what is it all about?”

2:25 – 2:50 pm
Jasmohan S. Bajaj, M.D.
Associate Professor
Internal Medicine, Gastroenterology
Virginia Commonwealth University, School of Medicine
“Gut Microbiota in Cirrhosis”

2:50 – 3:05 pm
Break

SESSION IV: Obesity & Nutrition
Moderators: Dr. Wu & Dr. Francis
MEC LH2
20 minutes presentation followed by 5 minutes discussion
3:05 – 3:30 pm  
**David E. Cohen, M.D., Ph.D.**  
Director of Hepatology, Brigham & Women’s Hospital  
Professor of Medicine & Health sciences & Technology  
Harvard Medical School  
Director, Harvard-MIT Division of Health Sciences & Technology  
Harvard Medical School  
“Acyl-CoA-Mediated Control of Thermogenesis: Therapeutic Implications for Obesity and the Metabolic Syndrome”

3:30 – 3:55 pm  
**Dawn Sears, M.D.**  
Associate Professor of Medicine, Baylor Scott & White Healthcare and Texas A&M Health Science Center  
“Your liver is what you drink: the good, the bad, the ugly – coffee, soda, and alcohol”

3:55 – 4:20 pm  
**Chaodong Wu, M.D., Ph.D.**  
Associate Professor  
Department of Nutrition & Food Science  
Texas A&M University  
“Pathophysiology of Fat Deposition and Inflammation In Obesity-associated NAFLD”

**Closing Remarks and adjournment:**  
Dr. Richard Erickson, M.D. Chief of Gastroenterology & Clinical Science Director of Baylor Scott & White Digestive Diseases Research Center; Scott & White Healthcare
Chandrashekhar Gandhi, Ph.D.
Cincinnati Children’s Hospital Medical Center
Augmenter of liver regeneration and alcoholic liver disease
BACKGROUND & AIMS: Liver cirrhosis due to chronic alcohol ingestion develops in up to 20% of alcoholics, molecular mechanisms of which are poorly understood. Recently, liver-specific deficiency of a protein named augmenter of liver regeneration (ALR) was found to induce steatosis that progresses to steatohepatitis and modest fibrosis. We sought to investigate whether chronic alcohol ingestion promotes excessive hepatic fibrosis in ALR-deficient mice.

METHODS: Liver-specific ALR-deficient and wild type (WT) mice were placed on 4% alcohol-supplemented or isocaloric diet for 4 weeks. Liver sections were examined for histolopathology. Parameters of steatosis and fibrosis were measured by quantitative reverse-transcription polymerase chain reaction, immunohistochemistry, immunoblotting and enzymatic assays.

RESULTS: Alcohol ingestion caused decrease in hepatic ALR in both WT and ALR-deficient mice. The gene expression levels of the enzymes of alcohol metabolism (alcohol dehydrogenase 1, acetaldehyde dehydrogenase 1 and cytochrome P450-E1) increased in WT mice but decreased in ALR-deficient mice. Consequently, hepatic acetaldehyde levels were high in alcohol-fed ALR-deficient mice. Whereas alcohol induced steatosis and mild inflammation in WT mice, ALR-deficient mice showed minimal steatosis but excessive hepatocellular injury, inflammation, prominent ductular proliferation, and robust fibrosis. There was significantly greater increase in the hepatic protein levels of inflammatory cytokine TNFα in alcohol-fed ALR-deficient mice but hepatoprotective IL6 and anti-inflammatory IL10 were lower than in WT mice. These pathological changes were accompanied by high oxidative stress and low GSH, robust lipid peroxidation, and mitochondrial DNA damage. Alcohol ingestion also induced hepatic accumulation of non-heme iron and decreased glutaredoxin 5 expression.

CONCLUSIONS: ALR deficiency or anomaly can be an important mechanism of the development of excessive liver fibrosis and other features of chronic alcoholic liver disease.

Satdarshan Monga, MD
University of Pittsburgh

Cell-Cell Junctions a Barriers: Function Beyond Cell Adhesion

Cell-cell junctions play an important role in maintenance of intercellular adhesion. These junctions include adherens junctions (AJ), desmosomes and tight junctions (TJ). Cell adhesion not only is important in physiological processes such as development, but its dysregulation is paramount in pathologies like cancers where it can contribute to tumor growth, local invasion and metastasis. In recent years another important function of these junctions has been realized and that is in the maintenance of barriers within a tissue. Examples of such phenomena are rampant in intestinal barriers, blood brain barrier and one of the lesser-understood blood bile barriers. Liver is a highly vascular organ allowing 1-2 liters of blood to pass through it every minute. Additionally hepatocytes produce bile and liver can generate anywhere between 250 ml-1 liter of bile every day. However, owing to the function of a barrier, blood and bile stay within distinct compartments in the liver such that blood is towards the basolateral or sinusoidal surface of hepatocyte, while bile, secreted from apical side of hepatocyte is channeled from that surface towards bile ductules. While endothelial cell-cell junctions play an important role in maintaining tissue barriers in many organs, liver is unique in that endothelial cells that line the blood containing sinusoids, lack traditional tight junctions and basement membrane for the purpose of free exchange of metabolites, toxicants and nutrients between blood and hepatocytes. What is known to maintain blood biliary barrier are the TJ situated close to apical surface of hepatocytes that allow these cells to adhere closely together preventing bile to trickle along lateral surface of hepatocytes. Recently, a loss of function mutation in a key TJ protein called ZO-2 or TJP2 was implicated in a subset of progressive familial intrahepatic cholestasis patients that are traditionally linked to mutations in bile acid transporters like ATP8B1, BSEP, MRP2. The function of AJ in hepatocytes has not been addressed although we have reported Claudin-2, a TJ protein to be regulated by β-catenin. Since β-catenin is a critical component of AJ where it links E-cadherin to Actin cytoskeleton, we generated hepatocyte-specific β-catenin knockouts (KO1). Intriguingly, AJ were maintained in these animals due to an upregulation of γ-catenin at AJ. We next conditionally knocked out both β-catenin and γ-catenin in the liver that led to notable morbidity such that most double
knockouts (DKO) die by 30 days after birth. These mice showed failure to thrive and severe jaundice confirmed by significant hyperbilirubinemia, elevated alkaline phosphatase and increased hepatic and serum bile acids. Histology too was reminiscent of PFIC and showed ductular reaction, fibrosis and inflammation. Analysis of PFIC cases also identified a small subset of patients which shows consistent loss of β-catenin and γ-catenin protein expression in liver. Thus, we report a novel function of AJ in regulating blood bile barrier, independent of TJ proteins. Further, a subset of PFIC cases could be due to aberrations in expression of AJ components and this observation could have implications in disease biology and therapeutics.

Huiping Zhou, PhD
Virginia Commonwealth University

**Conjugated bile acids promote cholangiocyte proliferation via activation of sphingosine-1-phosphate receptor 2**

Cholangiocytes are the major target cells in a number of human cholestatic liver diseases. Cholangiocytes are continuously exposed to high concentrations of bile salts. The bile duct obstruction is a potent stimulus for cholangiocyte proliferation. Bile acids have been reported to activate a number of intracellular signaling pathways including: PKC, PI3K, MAP Kinase, and ERK1/2. We have recently reported that conjugated bile acids (CBAs) activate the AKT and ERK1/2 signalling pathways via the G protein coupled receptor (GPCR) sphingosine-1-phosphate receptor 2 (S1PR2) in hepatocytes and cholangiocytes. However, the role of S1PR2 in CBA-mediated cholangiocyte proliferation has not been identified and is the focus of this study.

**Methods:** Human and mouse normal cholangiocyte cell lines and primary cholangiocytes isolated from wild type and S1PR2−/− mice were used for in vitro studies. Bile duct ligation (BDL) mouse models were used to examine the role of S1PR2 in cholangiocyte proliferation in vivo. The mRNA and protein levels of target genes were determined by real-time RT-PCR and Western blot analysis, respectively. Cell proliferation was determined using CCK-8 kit. The immunohistochemistry was used to examine the cholangiocyte proliferation in vivo.

**Results:** The S1PR2 is the predominant S1P receptor expressed in both human and mouse cholangiocytes. Both TCA- and S1P-induced cell migration and proliferation were inhibited by a specific shRNA and an antagonist of S1PR2 in cholangiocytes. BDL induced expression of S1PR2 in cholangiocytes. BDL-induced cholangiocyte proliferation and liver fibrosis were significantly reduced in S1PR2−/− mice.

**Discussion/Conclusion:** The levels of CBAs in serum and liver are significantly elevated in chronic cholestasis, which is correlated with bile duct obstruction. The results in this study suggestion that CBAs promote cholangiocyte proliferation by regulating the expression and activation of S1PR2. The CBA-mediated activation of the S1PR2 signaling pathways may represent novel therapeutic targets for cholestatic liver diseases.

Shrikant Anant, PhD
University of Kansas Medical Center

*TBD*

Robert Schwabe, MD
Columbia University
Hepatocellular carcinoma originates from hepatocytes but not from liver progenitor cells

Hepatocellular carcinoma (HCC) is the third leading cause of cancer mortality world-wide. Despite its nomenclature, the cellular origin of HCC remains elusive with hepatocytes and progenitor cells representing sources for newly generated hepatocytes in chronic liver injury and subsequently developing HCC. Determining the cellular source for HCC has high relevance for primary and secondary prevention strategies, as HCCs with progenitor signature carry a worse prognosis. To determine the cellular origin of HCC, we employed complementary cell fate tracing approaches to label progenitor cells and hepatocytes in murine hepatocarcinogenesis. Tumors arose from hepatocytes but not progenitor cells in genetic, genotoxic and dietary HCC models. Almost all A6-, AFP- and cytokeratin 19-positive cells within tumors but not in the surrounding liver were derived from hepatocytes. In summary, our findings suggest that hepatocytes are the main source for HCC in mice, and that progenitor signature does not reflect progenitor origin but rather de-differentiation of hepatocyte-derived tumor cells.

David Cohen, MD
Brigham & Women's Hospital/Harvard Medical School

Pradeep Dudja, PhD
University of Illinois

All-trans-retinoic acid (ATRA): A novel potential therapeutic agent for inflammatory diarrhea

Background: DRA (Down Regulated in Adenoma) or SLC26A3 is the major apical anion exchanger mediating Cl- absorption in intestinal epithelial cells (IECs). Reduction in DRA function and expression has been implicated in diarrhea associated with inflammatory bowel diseases (IBD). Upregulation of DRA, therefore, appears to be a novel approach to treat IBD associated diarrhea. In this regard, ATRA, a key metabolite of vitamin A is known to have anti-inflammatory and immunomodulatory properties. We earlier showed that ATRA, increased DRA function, expression and promoter activity through RAR-β via the involvement of transcription factor HNF-1β. Whether, ATRA could modulate DRA function and expression under inflammatory conditions is not known.

Aims: The aims were to evaluate the efficacy of ATRA in attenuating the inhibitory effects of IFN-γ on DRA utilizing Caco-2 cells as an in vitro model and Dextran sodium sulfate (DSS)-induced colitis as an in vivo mouse model.

Methods: Caco-2 cells grown on filter inserts were co-treated with IFN-γ (30 ng/ml) and ATRA for 24h or DSS colitis (3% DSS in drinking water for 7 days) mice were co-treated with ATRA (1 mg/kg body wt., i.p. for 7 days).

Results: Data demonstrated that ATRA abrogated IFN-γ induced decrease in DRA function as measured by DIDS-sensitive 125I uptake. Parallel to this, IFN-γ-induced decrease in DRA mRNA (50%, p<0.05) and protein (40%, p<0.05) was also markedly alleviated by ATRA. Further, ATRA significantly blocked the inhibitory effects of IFN-γ on DRA promoter activity. To evaluate if ATRA exerted these effects through modulation of IFN-γ induced signaling cascade, signal transducer and activator of transcription factor-1 (STAT-1) phosphorylation levels were analyzed. IFN-γ treatment induced the activation of STAT-1, however, ATRA co-treatment significantly diminished IFN-γ induced STAT-1 phosphorylation. In DSS- colitis, mouse model, ATRA treatment attenuated the reduced expression of DRA mRNA and protein levels in distal colon of DSS-mice. Further, the enhanced expression of inflammatory cytokines IL-1β (~10 fold) and CXCL1 (~18 fold) induced by DSS was also alleviated by ATRA treatment.

Conclusions: These data indicate that ATRA increases DRA function and expression under inflammatory conditions and this could serve as a novel therapeutic approach in IBD associated diarrhea.

Narendra Kumar, PhD
Texas A&M Health Science Center
Role of Janus Kinase 3 in Mucosal Differentiation & Predisposition to Colitis

Abstract: Janus kinase 3 (Jak3) is a non-receptor tyrosine kinase expressed in both hematopoietic and non-hematopoietic cells. Previously we characterized the functions of Jak3 in cytoskeletal remodeling, epithelial wound healing, and mucosal homeostasis. However, role of Jak3 in mucosal differentiation and inflammatory bowel disease was not known. In this report, we characterize the role of Jak3 in mucosal differentiation, basal colonic inflammation, and predisposition towards colitis. Using Jak3 knock out (KO) mice model, we show that Jak3 is expressed in colonic mucosa of mice and loss of mucosal expression of Jak3 resulted in reduced expression of differentiation markers for the cells of both enterocytic and secretory lineages. Jak3 KO mice showed reduced expression of colonic villin, carbonic anhydrase, secretory mucin muc2, and increased basal colonic inflammation reflected by increased level of pro-inflammatory cytokines IL-6 and IL-17A in colon along with increased colonic MPO activity. The inflammations in KO mice were associated with shortening of colon length, reduced caecum length, decreased crypt heights, and increased severity towards DSS-induced colitis. In differentiated human colonic epithelial cells, Jak3 redistributed to baso-lateral surfaces and interacted with adherens junction (AJ) protein β-catenin. Jak3 expression in these cells was essential for AJ localization of β-catenin and maintenance of epithelial barrier functions. Collectively these results demonstrate the essential role of Jak3 in colon where it facilitated mucosal differentiation through promoting the expression of differentiation makers, and enhanced colonic barrier functions through AJ localization of β-catenin.

Gut-feeling, what is it all about?

Chronic low-grade inflammation is a well-established characteristic of the several human diseases including IBD, obesity, and associated complications. Traditionally, such inflammation has been considered a product of metabolic deterioration mainly emanating from liver and adipose tissue. In addition to a role for liver and adipose tissue in the metabolic deterioration that is characteristic of obesity, our data suggest a key role for low-grade inflammation of intestinal mucosa as an early step in onset obesity and associated metabolic complications. We show that loss of mucosal expression of Janus kinase 3 (Jak3), a non-receptor tyrosine kinase, results in low-grade chronic inflammation in intestine and a tendency toward obesity. We have also identified the essential roles of Jak3 in IL-2-induced cytoskeletal remodeling, intestinal restitution, mucosal homeostasis, trans-molecular regulation of Jak3 activation and cancer therapy. Intestinal mucosa is the first tissue that interacts with dietary components and luminal microbiota both of which are known to regulate diabesity (diabetes+ obesity). Since intestinal mucosal response to these luminal contents are in-part regulated by toll-like receptors (TLRs), our data suggest a key role of Jak3 in the regulation of TLR activation-mediated mucosal tolerance not only towards luminal microbiota but also towards regulation of systemic circulated pro-inflammatory cytokines. Together these observations reinforce the age old saying that it’s the GUT and associated-fillings that is responsible for most of the inflammation associated complications.