WAKE FOREST
UNIVERSITY

GRADUATE SCHOOL *of*
ARTS & SCIENCES

Final examination of

Dipen A. Vyas

for the degree of

Doctor of Philosophy

COMMITTEE IN CHARGE

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Richard Dean Research Building
391 Technology Way
Conference Room 250
Thursday, April 16, 2015
2:00 p.m.

PROFESSOR(S) IN CHARGE OF RESEARCH
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FIELDS OF GRADUATE STUDY
Major Subject:
Physiology and Pharmacology

SUMMARY OF DISSERTATION
A THREE DIMENSIONAL MODEL OF HUMAN HEPATO-BILIARY ORGANOGENESIS

In recent years, tissue engineering strategies have emerged aimed at generating functional liver tissue by using decellularized liver scaffolds repopulated with liver cells. The quest continues for identifying the best cell source for liver tissue engineering, but one cell source in particular, human fetal liver progenitor cells (hFLPCs), presents the best chance of generating functional liver tissue because of their capability of differentiating into hepatocytes and cholangiocytes. To effectively use these cells for liver tissue engineering, it is critical to understand their developmental biology, ideally in an in vitro system. The goal of this thesis project was to generate a 3D in vitro model of liver tissue using tissue engineering strategies that can recapitulate fetal liver development. There are two key components to our in vitro model: acellular liver scaffold discs that provide 3D native liver tissue microarchitecture and microenvironment, and bipotent hFLPCs obtained from human fetal livers. When seeded with hFLPCs, the ECM discs formed 3D organoids like structure in culture over 3 weeks during the differentiation process. hFLPCs differentiated into both, hepatocytes and cholangiocytes within the organoids and displayed lineage specification and maturation process in a stepwise manner. Hepatocytes displayed transcriptional switch from α-fetoprotein to albumin and isoform switch of fetal cytochrome P450 3A7 to adult cytochrome P450 3A4. Functionally, the organoids displayed albumin and urea secretion along with phase 1 metabolism of diazepam and 7-ethoxy coumarin. The cholangiocytes formed bile duct structures by undergoing biliary morphogenesis process and attained apicobasal polarity in the organoids. The duct morphogenesis process was interrupted by inhibiting Notch signaling, thus creating a liver developmental disease model exhibiting a phenotype similar to Alagille syndrome. Altogether, these results describe, for the first time, a unique 3D in vitro model recapitulating hepato-biliary organogenesis. This model provides novel approaches for studying liver development, liver bioengineering, drug discovery and toxicology, and ultimately for treatment of liver disease.

SCHOLASTIC VITAE

EDUCATION

2015	Ph.D., Integrative Physiology and Pharmacology Wake Forest University, Winston-Salem, NC
2015	M.B.A., Wake Forest University School of Business Winston-Salem, NC
2009	M.S., Pharmaceutical Sciences Campbell University College of Pharmacy and Health Sciences, Buies Creek, NC
2007	B.S., Pharmacy North Gujarat University, School of Pharmacy, Mehsana, India

AWARDS

2014	Venture Capital Investment Competition, 2nd place, Southeast Regional, Georgetown University
2013	Wake Forest University Biotechnology Conference and Case Competition, 2nd
2012	Wake Forest University Research Day, Runner up – Translational Sciences Poster Presentation
2011	NC Tissue Engineering and Regenerative Medicine Society, Best Oral Presentation
2009 – Present	Graduate Fellowship, WFU Graduate School of Arts and Sciences

PUBLICATIONS

Manuscripts

- **Vyas DA**, Baptista PM, Moran E, Soker S, et.al. Self-Assembled Liver Organoids Recapitulate Hepato-Biliary Organogenesis In Vitro. In Preparation.
- Moran EC, Baptista PM, **Vyas D**, Riberio MH, Atala A, Sparks JL, Soker S. "A Bioengineered Liver Platform to Study Fluid Flow Regulation of Liver Tissue Organization". Submitted to Biomaterials
- Mokhtari S, Baptista PM, **Vyas D**, Freeman J, Moran E, Zabarsky Z, Porada C, Soker S, Almeida-Porada G. A New Approach to Expand Cord Blood Derived Hematopoetic Stem Cells Using Bioengineered Human Fetal Liver Tissue Constructs. In Preparation.

Book Chapters/Reviews

- Baptista P.M., **Vyas D**., Soker S., Liver Regeneration and Tissue Engineering, November 2010.
- Baptista P.M., **Vyas D**., Soker S., Liver Regeneration: the role of bioengineering, Progress in Molecular and Environmental Bioengineering- From Analysis and Modeling to Technology Applications (ISBN: 978-953-307-268-5), 1st Ed., Intech May 2011.
- Pedro M. Baptista, Emma Moran, **Dipen Vyas**, Thomas Shupe, Shay Soker The Liver Extra-Cellular Matrix and Stem/Progenitor Cells for Liver Regeneration "Regenerative Medicine Technology as Applied to Organ Transplantation", edited by Giuseppe Orlando, Elsevier, April 2013.
- **Dipen Vyas**, Emma Moran, Abritee Dhal, Pedro M. Baptista, Clinical Aspects of Regenerative Medicine: Hepatic system, "Translational Regenerative Medicine", edited by Anthony Atala and Julie Alickson, Elsevier, June 2013.

Review Articles

- Pedro M. Baptista, Dipen Vyas, Emma Moran, Zhan Wang, Shay Soker Human Liver Bioengineering Using a Whole Liver Decellularized Bioscaffold Methods in Molecular Biology, Volume on ³Organ Regeneration², Series Ed.: Walker, John M., ISSN: 1064-3745, 2012.
- Moran EC, Dhal A, Vyas D, Lanas A, Soker S, Baptista PM. "Whole-Organ Bioengineering: Current Tales of Modern Alchemy". Transl Res. 2014 Apr; 163(4):259-67. doi: 10.1016/j.trsl.2014.01.004

Podium Presentations

- **D. Vyas**, P.M. Baptista, E. Moran, S.Soker. Recapitulation of Human Hepato-Biliary Organogenesis in Self-Assembled Liver Organoid Culture. *TERMIS-AM, Washington D.C., Dec. 13-16th, 2014*
- **D. Vyas**, P.M. Baptista, S. Soker. Bioengineered Livers - A Platform for Drug Screening and Drug Toxicity Studies. *TERMIS-AM, Houston, Dec. 11-14th, 2011*
- **D. Vyas**, P.M. Baptista, S. Soker. Bioengineered Livers - A Platform for Drug Screening and Drug Toxicity Studies. *NCTERMS Annual Meeting, Winston-Salem, November 2011*
- **D. Vyas**, P.M. Baptista, S. Soker. Bioengineered Livers - The use of whole organ decellularization for the bioengineering of a human vascularized liver. *ACC Interdisciplinary Forum for Discovery in Life Sciences, Blacksburg, October 3-6, 2010.*