Section of Neonatology, Perinatal and Pulmonary Biology

Division Details

RESEARCH AND TRAINING DETAILS	
Faculty	**56**
Joint Appointment Faculty	**1**
Research Fellows and Post Docs	**13**
Research Graduate Students	**31**
Total Annual Grant Award Dollars	**$12,629,518**
Total Annual Industry Award Dollars	**$575,404**
Total Publications	**143**

CLINICAL ACTIVITIES AND TRAINING	
Staff Physicians	**34**
Clinical Fellows	**15**

Row 1: A Perl, S Merhar, S Kallapur, T Weaver, L Muglia, J Whitsett, J Greenberg, B Haberman, A Nathan, K Wedig, Y Xu

Row 2: D Sinner, T Cahill, M Pavlicev, P Arumugam, K Wikenheiser-Brokamp, T Kalin, B Poindexter, Y Maedea, H Deshmukh, V Narendran, D Swarr, S Bhattacharyya

Row 3: J Shannon, J Bridges, V Kalinichenko, S Ahlfeld, B Trapnell, A Jobe, E Hall, T Suzuki, P Kingma

Research Highlights

SINCERA: A Pipeline for Single-Cell RNA-Seq Profiling Analysis

A major challenge in developmental biology is to understand the genetic and cellular processes/programs driving organ formation, and differentiation of the diverse cell types that comprise the embryo. While recent studies using single cell transcriptome analysis illustrate the power to measure and understand cellular heterogeneity in complex biological systems, processing large amounts of RNA-seq data from heterogeneous cell populations creates the need for readily accessible tools for the analysis of single-cell RNA-seq (scRNA-seq) profiles. The present study presents a generally applicable analytic pipeline (SINCERA: a computational pipeline for SINgle CEll RNA-seq profiling Analysis) for processing scRNA-seq data from a whole organ or sorted cells. The pipeline supports the analysis for: 1) the distinction and identification of major cell types; 2) the identification of cell type specific gene signatures; and 3) the determination of driving forces of given cell types. We applied this pipeline to the RNA-seq analysis of single cells isolated from embryonic mouse lung at E16.5. Through the pipeline analysis, we distinguished major cell types of fetal mouse lung, including epithelial, endothelial, smooth muscle, pericyte, and fibroblast-like cell types, and identified cell type specific gene signatures, bioprocesses, and key regulators. Implemented in R, and licensed under the GNU General Public License v3, SINCERA is freely available from Cincinnati Children's PBGE website.

Metagenomic Sequencing with Strain-Level Resolution Implicates Uropathogenic E. coli in Necrotizing Enterocolitis and Mortality in Preterm Infants

Necrotizing enterocolitis (NEC) afflicts approximately 10% of extremely preterm infants with high fatality. There is an implication of inappropriate bacterial colonization with Enterobacteriaceae, but there has been no specific pathogen identified. We identify uropathogenic E. coli (UPEC) colonization as a significant risk factor for the development of NEC and subsequent mortality. We describe a large-scale deep shotgun metagenomic sequence analysis of the early intestinal microbiome of 144 preterm and 22 term infants. Using a pan-genomic approach to functionally subtype the E. coli, we identify genes associated with NEC and mortality that indicate colonization by UPEC. Metagenomic multilocus sequence typing analysis further defined NEC-associated strains as sequence types often associated with urinary tract infections, including ST69, ST73, ST95, ST127, ST131, and ST144. Although other factors associated with prematurity may also contribute, this report suggests a link between UPEC and NEC, and indicates the need for further attention to these sequence types as potential causal agents.

Functional and Structural Connectivity of the Visual System in Infants with Perinatal Brain Injury

In this study, we evaluated term and preterm infants with perinatal brain injury and term controls in the first 8 weeks of life using task-based functional MRI, functional connectivity during a visual task, and structural connectivity using diffusion tensor imaging. We found that infants with brain injury had reduced functional and structural connectivity compared to term control infants. Specifically, infants with brain injury had reduced activation in the expected area of the occipital cortex, weaker connectivity between visual areas and other areas of the brain during the visual task, and reduced fractional anisotropy (a measure of white matter integrity) in white matter tracts projecting to visual regions. Our next steps will be to correlate these early neuroimaging findings with later visual outcomes in this cohort.

Significant Publications

Cai Y, **Bolte C**, Le T, Goda C, **Xu Y, Kalin TV, Kalinichenko VV**. **FOXF1 maintains endothelial barrier function and prevents edema after lung injury**. *Sci Signal*. 2016 Apr 19;9(424):ra40.

Multiple signaling pathways, structural proteins, and transcription factors are all involved in the regulation of endothelial barrier function. The forkhead protein FOXF1 is a key transcriptional regulator of embryonic lung development, and we used a conditional knockout approach to examine the role of FOXF1 in adult lung homeostasis, injury, and repair. Tamoxifen-regulated deletion of both Foxf1 alleles in endothelial cells of adult mice (Pdgfb-iCreER/Foxf1(-/-)) caused lung inflammation and edema, leading to respiratory insufficiency and death. Deletion of a single Foxf1 allele made heterozygous Pdgfb-iCreER/Foxf1(+/-)mice more susceptible to acute lung injury. Findings showed decreased FOXF1 abundance in pulmonary endothelial cells of human patients with acute lung injury. Gene expression analysis of pulmonary endothelial cells with homozygous FOXF1 deletion indicated reduced expression of genes critical for maintenance and regulation of adherens junctions. FOXF1 knockdown in vitro and in vivo disrupted adherens junctions, enhanced lung endothelial permeability, and increased the abundance of the mRNA and protein for sphingosine 1-phosphate receptor 1 (S1PR1), a key regulator of endothelial barrier function. Chromatin immunoprecipitation and luciferase reporter assays demonstrated that FOXF1 directly bound to and induced the transcriptional activity of the S1pr1 promoter. Pharmacological administration of S1P to injured Pdgfb-iCreER/Foxf1(+/-)mice restored endothelial barrier function, decreased lung edema, and improved survival. Thus, FOXF1 promotes normal lung homeostasis and repair, in part, by enhancing endothelial barrier function through activation of the S1P/S1PR1 signaling pathway.

Zhang G, Bacelis J, **Lengyel C**, **Teramo K**, Hallman M, Helgeland O, Johansson S, Myhre R, Sengpiel V, Njolstad PR, Jacobsson B, **Muglia L**. **Assessing the Causal Relationship of Maternal Height on Birth Size and Gestational Age at Birth: A Mendelian Randomization Analysis**. *PLoS Med*. 2015 Aug 18;12(8):e1001865.

Observational epidemiological studies indicate that maternal height is often associated with gestational age at birth and fetal growth measures (i.e., shorter mothers deliver infants at earlier gestational ages with lower birth weight and birth length). To explain these associations, postulating of different mechanisms has occurred. This study aimed to investigate the casual relationships behind the strong association of maternal height with fetal growth measures (i.e., birth length and birth weight) and gestational age by a Mendelian randomization approach. Our results demonstrate that the observed association between maternal height and fetal

growth measures (i.e., birth length and birth weight) is mainly defined by fetal genetics. In contrast, the association between maternal height and gestational age is more likely to be causal. In addition, our approach that utilizes the genetic score derived from the nontransmitted maternal haplotype as a genetic instrument is a novel extension to the Mendelian randomization methodology in casual inference between parental phenotype (or exposure) and outcomes in offspring.

Division Publications

1. Acciani TH, Suzuki T, Trapnell BC, Le Cras TD. **Epidermal Growth Factor Receptor Signalling Regulates Granulocyte-Macrophage Colony-Stimulating Factor Production by Airway Epithelial Cells and Established Allergic Airway Disease.** *Clin Exp Allergy*. 2016; 46:317-28.
2. Adler J, Saeed SA, Eslick IS, Provost L, Margolis PA, Kaplan HC. **Appreciating the Nuance of Daily Symptom Variation to Individualize Patient Care.** *EGEMS (Wash DC)*. 2016; 4:1247.
3. Agricola ZN, Jagpal AK, Allbee AW, Prewitt AR, Shifley ET, Rankin SA, Zorn AM, Kenny AP. **Identification of Genes Expressed in the Migrating Primitive Myeloid Lineage of Xenopus Laevis.** *Dev Dyn*. 2016; 245:47-55.
4. Ahlfeld SK, Wang J, Gao Y, Snider P, Conway SJ. **Initial Suppression of Transforming Growth Factor-Beta Signaling and Loss of Tgfbi Causes Early Alveolar Structural Defects Resulting in Bronchopulmonary Dysplasia.** *Am J Pathol*. 2016; 186:777-93.
5. Althabe F, Thorsten V, Klein K, McClure E, Hibberd P, Goldenberg R, Carlo W, Garces A, Patel A, Pasha O. **The Antenatal Corticosteroids Trial (Act)'S Explanations for Neonatal Mortality - a Secondary Analysis.** *Reprod Health*. 2016; 13:62.
6. American Academy Of Pediatrics Committee On Fetus And Newborn ACOOAGCOOP. **The Apgar Score.** *Pediatrics*. 2015; 136:819-22.
7. Arnett MG, Muglia LM, Laryea G, Muglia LJ. **Genetic Approaches to Hypothalamic-Pituitary-Adrenal Axis Regulation.** *Neuropsychopharmacology*. 2016; 41:245-60.
8. Arumugam PI, Mullins ES, Shanmukhappa SK, Monia BP, Loberg A, Shaw MA, Rizvi T, Wansapura J, Degen JL, Malik P. **Genetic Diminution of Circulating Prothrombin Ameliorates Multiorgan Pathologies in Sickle Cell Disease Mice.** *Blood*. 2015; 126:1844-55.
9. Assaf S, Chang D, Tiller C, Kisling J, Case J, Mund J, Slaven J, Yu Z, Ahlfeld S, Poindexter B. **Lung Parenchymal Development in Premature Infants without Bronchopulmonary Dysplasia.** *Pediatr Pulmonol*. 2015; 50:1313-19.
10. Bancalari E, Jain D, Jobe AH. **Prevention of Bronchopulmonary Dysplasia: Are Intratracheal Steroids with Surfactant a Magic Bullet?** *Am J Respir Crit Care Med*. 2016; 193:12-3.
11. Benitz WE, Committee on Fetus Newborn American Academy of Pediatrics. **Patent Ductus Arteriosus in Preterm Infants.** *Pediatrics*. 2016; 137:e20153730-e30.
12. Brandt EB, Biagini Myers JM, Acciani TH, Ryan PH, Sivaprasad U, Ruff B, LeMasters GK, Bernstein DI, Lockey JE, LeCras TD, Khurana Hershey GK. **Exposure to Allergen and Diesel Exhaust Particles Potentiates Secondary Allergen-Specific Memory Responses, Promoting Asthma Susceptibility.** *J Allergy Clin Immunol*. 2015; 136:295-303 e7.
13. Cai Y, Bolte C, Le T, Goda C, Xu Y, Kalin TV, Kalinichenko VV. **Foxf1 Maintains Endothelial Barrier Function and Prevents Edema after Lung Injury.** *Sci Signal*. 2016; 9:ra40.
14. Cortezzo DE, Sanders MR, Brownell EA, Moss K. **End-of-Life Care in the Neonatal Intensive Care Unit: Experiences of Staff and Parents.** *Am J Perinatol*. 2015; 32:713-24.
15. Coya JM, Akinbi HT, Saenz A, Yang L, Weaver TE, Casals C. **Natural Anti-Infective Pulmonary Proteins: In Vivo Cooperative Action of Surfactant Protein Sp-a and the Lung Antimicrobial Peptide Sp-B-N.** *J Immunol*. 2015; 195:1628-36.

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21. Du Y, Guo M, Whitsett J, Xu Y. **'Lunggens': A Web-Based Tool for Mapping Single-Cell Gene Expression in the Developing Lung.** *Thorax*. 2015; 70:1092-94.

22. Eichenwald EC, Committee on Fetus Newborn American Academy of Pediatrics. **Apnea of Prematurity.** *Pediatrics*. 2016; 137:e20153757-e57.

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Grants, Contracts, and Industry Agreements

Annual Grant Award Dollars

Investigator	Title	Sponsor	ID	Dates	Amount
James Patrick Bridges, PHD	Role of GPR116 in the Regulation of Alveolar Surfactant Pool Size	American Heart Association	13SDG17090028	7/1/2013 - 6/30/2017	$77,000
James Patrick Bridges, PHD	Role of GPR116 in Alveolar Homeostasis	National Institutes of Health	R01 HL131634	3/18/2016 - 2/28/2021	$402,550
Hitesh Deshmukh, MD-PHD	Role of Commensal Bacteria in Regulating Neutrophil-mediATED Host Defense in Neonates	National Institutes of Health	K08 HD084686	8/1/2015 - 5/31/2019	$259,740
Neera Goyal, MD	Preventive Health Services and ED Utilization among At-Risk Infants	Health Resources & Services Admin	R40MC29447	4/1/2016 - 3/31/2017	$100,000
James M Greenberg, MD	Cradle Cincinnati	City of Cincinnati	5582015340	6/17/2015 - 10/31/2016	$250,000
James M Greenberg, MD	Healthy Start Cincinnati	Health Resources & Services Admin	H49MC27823	9/1/2014 - 5/31/2019	$674,337
Margaret K Hostetter, MD	Child Health Research Career	National Institutes of	K12 HD028827	12/1/2015 -	$364,467

	Development Award (K12)	Health		11/30/2016	
Alan Hall Jobe, MD-PHD	Antenatal Steroid Treatment Strategies for Low Resource Countries	Bill & Melinda Gates Foundation	OPP1132910	10/26/2015 - 9/30/2017	$364,047
Alan Hall Jobe, MD-PHD	Initiation and Progression of Preterm Lung Injury with Ventilation	National Institutes of Health	R01 HD072842	6/1/2016 - 5/31/2017	$282,783
Tatiana Viktorovna Kalin, MD-PHD	Transcriptional Regulation of Pulmonary Fibrosis	National Institutes of Health	R56 HL126660	9/8/2015 - 8/31/2016	$390,000
Tatiana Viktorovna Kalin, MD-PHD	Transcriptional Regulation of Cancer Progression and Metastasis by Foxm1	American Cancer Society National	RSG1332501	7/1/2013 - 6/30/2017	$180,000
Vladimir V Kalinichenko, MD-PHD	Foxf1 Transcription Factor in Development of Pulmonary Capillaries	National Institutes of Health	R01 HL084151	5/8/2015 - 4/30/2019	$390,000
Vladimir V Kalinichenko, MD-PHD	Transcriptional Regulation of Goblet Cell Metaplasia	National Institutes of Health	R01 HL123490	8/5/2014 - 6/30/2018	$384,150
Paul Kingma, MD-PHD	Intestinal Motility and Gastroschisis	The Gerber Foundation	1557-3464	7/1/2013 - 6/30/2017	$57,443
Timothy D LeCras, PHD	Identification of Biomarkers for Patients with Generalized Lymphatic Anomaly, Kaposiform Lymphangiomatosis in Gorham Stout Disease	The Lymphatic Malformation Institute	LeCras, Tim, LMI	5/1/2015 - 4/30/2017	$149,725
Yutaka Maeda, PHD	Dissecting Tumor Heterogeneity in KRAS-Mutant Lung Cancer	American Lung Association	RG309608	8/1/2014 - 7/31/2016	$40,000
Kristin R Melton, MD Eric Kirkendall, MD	Improving Intensive Care Medication Safety through EHR-based Algorithms	National Institutes of Health	R01 LM012230	9/15/2015 - 8/31/2019	$174,656
Stephanie L Merhar, MD	Serial Neuroimaging in Infants with Necrotizing Enterocolitis	Cerebral Palsy International Research Fd	EH01400	1/1/2015 - 12/31/2015	$75,000
Ardythe Morrow, PHD	Ctr for Disease Control and Prevention	Ctr for Disease Control and Prevention	2002015M63224	7/14/2015 - 2/14/2017	$31,781
Ardythe Morrow, PHD	DNA Attenuates Inflammatory Responses through Altering RAGE Signaling	National Institutes of Health (The Research Instit at Nationwide Hosp)	R01 AT006880	7/1/2012 - 6/30/2016	$17,786
Louis Muglia	March of Dimes Prematurity Research Center Ohio Collaborative	March of Dimes	22-FY16-125	1/1/2016 - 12/31/2016	$2,000,000

Louis Muglia	Systems Biology Approaches to Birth Timing and Preterm Birth Risk	Bill & Melinda Gates Foundation	OPP1113966	11/17/2014 - 10/31/2016	$645,295
Louis Muglia	Preventing Prematurity: Establishing a Network for Innovation and Discovery	Bill & Melinda Gates Foundation	OPP1151188	4/20/2016 - 12/31/2016	$50,000
Louis Muglia;Claire A Chougnet, PHD	Maternal Temperament, Stress, and Inflammation in Preterm Birth	National Institutes of Health	R01 HD078127	9/1/2013 - 8/31/2017	$310,298
Laurie A Nommsen-Rivers, PHD	Improving Lactation Success in Pre-diabetic Mothers	National Institutes of Health (University of Cincinnati)	K12 HD051953	7/1/2014 - 6/30/2016	$108,000
Anne Karina Perl, PHD	Matrix Fibroblasts are required for Alveolar Homeostasis and regrowth	National Institutes of Health	R56 HL123969	9/8/2015 - 8/31/2016	$390,000
Brenda Poindexter, MD-MS	Gastrin-Releasing Peptide and Bronchopulmonary Dysplasia	National Institutes of Health (Duke University)	R01 HL105702	10/1/2014 - 7/31/2016	$62,424
Brenda Poindexter, MD-MS	NICHD Cooperative Multicenter Neonatal Research Network	National Institutes of Health	UG1HD027853	4/1/2016 - 3/31/2021	$294,601
Debora Ines Sinner, PHD	Molecular Mechanisms Underlying Upper Airway Patterning and Tracheomalcia	National Institutes of Health	K01 HL115447	8/1/2012 - 7/31/2016	$112,397
Bruce C Trapnell, MD	Childhood Interstitial Lung Disease (chILD) and Chemokine (C-C motif) Receptor 2 (CCR2) Deficiency	American Thoracic Society (Johns Hopkins School of Medicine)	PBMCCCR2	10/31/2013 - 10/30/2015	$4,847
Bruce C Trapnell, MD	Macrophage Based Gene Therapy for Hereditary Pulmonary Alveolar Proteinosis	National Institutes of Health	R01 HL118342	5/1/2014 - 4/30/2018	$664,101
Bruce C Trapnell, MD	RLDC: Molecular Pathway-Driven Diagnostics & Therapeutics for Rare Lung Disease	National Institutes of Health	U54 HL127672	9/18/2014 - 7/31/2019	$843,384
Timothy E Weaver, PHD	Stard7, a Novel Inhibitor of Allergic Lung Disease	National Institutes of Health	R01 HL122130	1/1/2014 - 12/31/2017	$390,000
Scott Lewis Wexelblatt	Neonatal Abstinance Syndrome (NAS) Project	Nationwide Children's Hospital (University Hospitals)		7/1/2012 - 12/31/2016	$61,443
Jeffrey A Whitsett, MD	Omics of Lung Diseases	National Institutes of	K12 HL119986	9/1/2013 -	$269,603

		Health		5/31/2016	
Jeffrey A Whitsett, MD	Transcriptional Programming of Asthma Related Pathology in Respiratory Epithelia	National Institutes of Health	R01 HL095580	4/1/2016 - 3/31/2018	$528,560
Jeffrey A Whitsett, MD Bruce C Trapnell, MD Francis McCormack	Lung and Cardiovascular Development and Disease Pathogenesis Training Program	National Institutes of Health	T32 HL007752	7/1/2014 - 6/30/2019	$147,493
Jeffrey A Whitsett, MD	Airway Progenitor Cell Proliferation and Differentiation During Lung Repair	National Institutes of Health	U01 HL110964	1/1/2016 - 12/31/2016	$639,744
Jeffrey A Whitsett, MD	Molecular Atlas of Lung Development - Data Coordinating Center	National Institutes of Health (Duke University)	U01 HL122638	6/15/2014 - 4/30/2019	$50,881
Jeffrey A Whitsett, MD S Steven Potter, PHD	Lung MAP Atlas Research Center	National Institutes of Health	U01 HL122642	6/15/2014 - 4/30/2019	$390,982
Total Annual Grant Award Dollars					**$12,629,518**

Annual Industry Award Dollars

Investigator	Industry Sponsor	Amount
Paul Kingma, MD-PHD	Airway Therapeutics & Steve Linberg	$69,300
Vivek Narendran, MD	Chiesi	$181,104
Jeffrey A Whitsett, MD	Transcriptx, Inc.	$325,000
Total Annual Industry Award Dollars		**$575,404**