<table>
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<tr>
<th>Type of research application</th>
<th>T2</th>
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<tbody>
<tr>
<td>Title of project</td>
<td>The Clinical Effectiveness of Novel Molecular Assays to Diagnose Osteoarticular Infections in Children</td>
</tr>
</tbody>
</table>
| Applicant Names (s) | Dr. Larry J. Anderson, M.D.  
Professor and Marcus Chair of Infectious Diseases  
Division of Pediatric Infectious Diseases  
Department of Pediatrics, Emory University, Atlanta  
Larry.anderson@emory.edu  
Dr. Jill C. Flanagan, M.D.  
Pediatric Surgeon  
Children's Orthopaedics of Atlanta/Children's Healthcare of Atlanta  
Jflanagan@childrensortho.com  
Dr. Craig A. Shapiro, M.D.  
Pediatrician, Infectious Diseases Specialty  
Children's Orthopaedics of Atlanta/Children’s Healthcare of Atlanta  
Emory University, Atlanta  
Cshapi2@emory.edu  
Dr. Gregory Gibson, PhD.  
Professor and Director of Center for Integrative Genomics  
Georgia Institute of Technology, Atlanta  
greg.gibson@biology.gatech.edu  
Dr. Robert Jerris, Ph.D.  
Director, Clinical Microbiology  
Scottish Rite and Egleston Children's Hospital, Emory University, Atlanta  
Robert.Jerris@emory.edu  
Dr. Lars Westblade, Ph.D., Clinical Microbiologist  
Scottish Rite and Egleston Children's Hospital  
Emory University, Atlanta  
Lars.westblade@emory.edu  
Dr. Samadhan Jadhao, D.V.M., PhD  
Research Fellow  
Division of Pediatric Infectious Diseases  
Department of Pediatrics, Emory University, Atlanta  
Samadhan.Jadhao@emory.edu |
| Requested award amount (Direct Cost) | US $ 50000 for two years (US $ 25000/year for two years) |
Abstract

Osteoarticular infections of bones and joints caused by pathogenic bacteria pose a significant risk to health and mobility of young children. Rapid identification of the causative bacterial pathogen is important to provide optimal treatment and minimize the risk of complications rendered by ineffective therapy. For example, without identification of the causative organism the clinician is forced to use a broad-spectrum antibiotic which increases the risks for side effects including clostridium difficile colitis. Epidemiologic studies of osteoarticular infections show that the causative organism is detected in 50-60% of cases when tissue, synovial fluid, and blood are obtained and cultured. Thus, no organism is identified in up to 50% of cases by culture methods despite availability of specimens. With culture the most commonly detected bacteria in osteoarticular infections in children are *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Streptococcus pneumoniae*. Less is known about fastidious organisms often missed by culture. With molecular detection, we are finding other bacteria such as *Kingella kingae*, a gram negative coccobacillus, in osteoarticular infections. With PCR, *Kingella kingae* has been detected in up to 45% of cases of bone and joint infections in preschool children. The use of a rapid and sensitive PCR assays to detect *Kingella kingae*, other fastidious bacteria, and other difficult to culture specimens, e.g. from patients with prior antibiotic treatment, should improve our ability to detect the cause and manage osteoarticular infections in children. Another challenge in managing these infections is antibiotic resistance. Since bacterial genes associated with antimicrobial resistance have been identified, PCR assays can detect their presence and guide choice of antibiotic therapy. We propose to use pathogen-specific and resistance gene-specific real time PCR assays to detect the bacteria and antibiotic resistance in specimens from children with osteoarticular infections. The resulting data will determine frequency that PCR diagnostics detect bacteria otherwise missed and help provide optimal antibiotic therapy. These data will be used to determine the potential cost savings, patient safety benefits, and increase in treatment efficacy that PCR testing can provide. Finally, since testing for common bacterial and resistance genes will require multiple different PCR assays, we will adapt the individual assays to the Fluidigm microfluidic PCR system. This system provides an easy and efficient way to run multiple PCR assays in parallel. We will compare the sensitivity and specificity of this system to individual real time PCR assays. This proposal is based on collaborations among physicians and scientists at Children’s Healthcare of Atlanta (CHOA) at Scottish Rite and Egleston, Department of Pediatrics at CHOA and Emory University School of Medicine, and the Center for Integrative Genomics, Georgia Institute of Technology.
BIOGRAPHICAL SKETCH

NAME
Larry J. Anderson

POSITION TITLE
Professor of Pediatrics, Marcus Chair of Infectious Diseases Co-Director, Pediatric Infectious Diseases
Emory University School of Medicine

eRA COMMONS USER NAME (credential, e.g., agency login)
LJANDER

EDUCATION/TRAINING

<table>
<thead>
<tr>
<th>INSTITUTION AND LOCATION</th>
<th>DEGREE (if applicable)</th>
<th>MM/YY</th>
<th>FIELD OF STUDY</th>
</tr>
</thead>
<tbody>
<tr>
<td>St. Olaf College, Northfield, MN</td>
<td>BA</td>
<td>05/1969</td>
<td>Chemistry</td>
</tr>
<tr>
<td>Harbor Medical School, Boston, MA</td>
<td>BA</td>
<td>06/1974</td>
<td>Medicine</td>
</tr>
<tr>
<td>Harbor General Hospital, Los Angeles, CA</td>
<td>MD</td>
<td>06/1977</td>
<td>Infectious Diseases</td>
</tr>
<tr>
<td>Beth Israel Hospital, Boston, MA</td>
<td>Board Cert.</td>
<td>06/1982</td>
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</table>

A. Personal Statement.

I have been involved in a variety of studies related to the epidemiology, diagnosis, and characterization of respiratory virus infection, vaccine development, and pathogenesis of respiratory syncytial virus (RSV) disease. Included in the epidemiologic studies, are those of surveillance, disease burden, and risk factors for RSV infection and studies of the immune response to a variety of infections including RSV. Our studies of the pathogenesis of RSV have been focused on vaccine development and especially the G gene and its chemokine motif. The G protein has a chemokine motif, CX3C, allows it to bind to the corresponding receptor, CX3CR1, and mimic some activities of the one CX3C chemokine, fractalkine. In studies in BALB/c mice and cells, we have demonstrated that this binding facilitates infection, is associated with chemokine-like induction of leukocyte migration, depresses the respiratory rate, participates in enhanced inflammatory response after FI-RSV vaccination, likely induces substance P, and modulates the cellular response to infection. More recently we have shown that monoclonal antibodies that bind to G and block binding to CX3CR1 decrease virus replication (Fc receptor mediated) and the inflammatory response (independent of the Fc receptor). These monoclonal antibodies also decrease the enhanced inflammatory response to infection in FI-RSV immunized mice. We are continuing to work on the role of the G protein in pathogenesis of RSV disease, as a potential target for immune or anti-viral drug treatment, and as, outlined in this proposal, the pathogenesis of FI-RSV enhanced disease and the human memory response to RSV.

B. Positions, Honors, and Awards

Positions and Employment
1977-1980: Medical Epidemiologist, Respiratory and Special Pathogens Branch, Viral Diseases Division, Bureau of Epidemiology, CDC, Atlanta, GA
1980-1982: Infectious Disease Fellow, Beth Israel Hospital, Boston, MA
1982-2006: Chief Respiratory and Enteric Viruses Branch, Division of Viral and Rickettsial Diseases, Center for Infectious Diseases, CDC, Atlanta, GA
2006-2010: Director, Division of Viral Diseases, National Center for Immunization and Respiratory Diseases, Centers for Disease Control and Prevention (CDC), Atlanta, GA
2010–present: Professor and Marcus Chair of Pediatric Infectious Diseases, Co-Director, Division of Infectious Diseases, Department of Pediatrics, Emory University School of Medicine, Atlanta, GA

Society Memberships
American Society for Microbiology
American Society for Virology
Fellow, Infectious Disease Society of America
Fellow, American Association for the Advancement of Science

**Patents Filed/Awarded**
Ralph A. Tripp, Michael P. Brown, Larry J. Anderson. CD40 ligand adjuvant for respiratory syncytial virus, 2000
David Ku, Larry J. Anderson. Method to Collect Specimens from the Lung in a Fashion that Optimizes the Amount of Lung Specimen and Eliminates Contamination from the Upper Respiratory Tract, 2008
Larry J. Anderson, Ralph A. Tripp, Lia Haynes. Anti-RSV Immunogens and Methods of Immunization, 2009

**C. Selected Peer-reviewed Publications (15 of 225)**
10 Panozzo CA, Stockman LJ, Curns AT, Anderson LJ. Use of respiratory syncytial virus surveillance data to optimize the timing of immunoprophylaxis. Pediatrics 2010;126:e116-23


D. Research Support

Ongoing

1U19AI095227
NIH NIAID
$250,000/yr
(Peebles, Program PI; Project PI, Moore) 7/1/2011 – 6/30/2016
“Host and Viral Determinants of Infant and Childhood Allergy and Asthma”
This project will define associations between RSV genotypes, bronchiolitis severity, inflammatory markers, and asthma development in infants. The study will also define pathogenic mechanisms of RSV clinical isolates
Role: Co-Investigator (1.2 months)

2012-N-14280
Centers for Disease Control and Prevention
$62,000/yr
2012-2017
Herpes Simplex Virus Type 1 and 2 Antibody Testing for NHANES 2012-2017
PI (1.2 months)

Completed

Center for Nanomedicine (Georgia Institute of Technology, Emory University, Children’s Hospital of Atlanta)
Seed Grant ($60,000) 2011-2013
SERS Assays for RSV and Influenza Detection
Co-PI with Ralph Tripp

HIP-ACTSI Seed Grants in Healthcare Innovation (Georgia Institute of Technology and Emory University)
Seed Grant ($25,000) 2011-2013
Validation of a device to diagnose pneumonia from coughed droplets
PI

CRADA with Trellis Bioscience, Inc.  Anderson (PI)  2012-2013
“Comparison of a G protein and an F protein monoclonal antibody for treatment of RSV disease”
Role: PI (2.5%)

Comparative Effectiveness Research: Evaluation of Innovative Diagnostic Technologies and/or Medical Devices (Georgia Tech and Children’s Healthcare of Atlanta)
2013 Seed Grant ($50,000)  2012-2013
“Non-invasive Diagnosis of Airway Resistance”
Co-I

5 R01 AI069275-04  Tripp (PI)  2006-2010
NIH/NIAID
“Antibody inhibition of respiratory syncytial virus G protein activity”
The major goals of this project are to develop vaccine candidates which generate antibody that blocks RSV G protein CX3C binding to CX3CR1 and prevents disease pathogenesis.
Role: Co-Investigator

CRADA with Trellis Bioscience, Inc.  Anderson (PI)  2007-2010
“Develop G protein monoclonal antibodies for treatment of RSV disease”
Role: Principal Investigator

Gates Foundation  Anderson (PI)  2009-2010
“Estimate mortality associated with RSV infection in Bangladesh”
The main goal is to estimate the contribution of Respiratory Syncytial Virus (RSV) to mortality associated with acute lower respiratory tract illness in developing countries and to determine the need for development of a safe and effective RSV vaccine.
Role: Principal Investigator
### BIOGRAPHICAL SKETCH

Provide the following information for the key personnel in the order listed on Form Page 2. 
Follow this format for each person. **DO NOT EXCEED TWO PAGES.**

<table>
<thead>
<tr>
<th>NAME</th>
<th>POSITION TITLE</th>
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<tbody>
<tr>
<td>Jill C. Flanagan, MD</td>
<td>Pediatric Orthopaedic Surgeon</td>
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</tbody>
</table>

**EDUCATION/TRAINING** *(Begin with baccalaureate or other initial professional education, such as nursing, and include postdoctoral training.)*

<table>
<thead>
<tr>
<th>INSTITUTION AND LOCATION</th>
<th>DEGREE (if applicable)</th>
<th>YEAR(s)</th>
<th>FIELD OF STUDY</th>
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<tbody>
<tr>
<td>The George Washington University</td>
<td>BS</td>
<td>June 2000</td>
<td>Biology</td>
</tr>
<tr>
<td>The George Washington University</td>
<td>MD</td>
<td>June 2004</td>
<td>Medicine</td>
</tr>
<tr>
<td>Children's Healthcare of Atlanta, Scottish Rite</td>
<td></td>
<td>2009-2010</td>
<td>Pediatric Orthopaedic Surgery</td>
</tr>
<tr>
<td>Certified Principal Investigator</td>
<td>CPI</td>
<td>2010</td>
<td></td>
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</table>

**EMPLOYMENT**

- **Orthopaedic Surgery Resident, July 2004 – June 2009**
  - The George Washington University; Washington, DC
  - Residency Director: Robert Neviaser, MD
- **Pediatric Orthopaedic Surgery Fellow, August 2009- July 2010**
  - Children’s Healthcare of Atlanta at Scottish Rite; Atlanta, GA
  - Fellowship Director: Michael Busch, MD
- **Pediatric Orthopaedic Surgeon – Children’s Orthopaedics of Atlanta, Sept 2010 – present**
  - Full time orthopaedic surgeon for a busy private practice
  - Hospital Affiliation: Children’s Healthcare of Atlanta

**HONORS/AWARDS**

- Ruth Jackson Orthopaedic Society Practice Enhancement Travelling Fellowship, 2011
- American Orthopaedic Association Emerging Leader, 2010

**PROFESSIONAL SOCIETIES**

- American Academy of Orthopaedic Surgeons (AAOS), 2004-present
- Ruth Jackson Orthopaedic Society (RJOS) 2004-present
- The American Society for Bone and Mineral Research (ASBMR) 2011-present
- Pediatric Orthopaedic Society of North America (POSNA) 2012-present
- Limb Lengthening and Reconstruction Society (LLRS) 2013-present
- Georgia Orthopaedic Society 2013 - present
- American Association of Pediatrics (AAP) 2013-present
• PUBLICATIONS
  • ORIGINAL RESEARCH
  • NON-EXPERIMENTAL ARTICLES
  • POSTERS AND PRESENTATIONS
    o Flanagan JC, Thornton RK, Greenbaum VJ, and Schmitz ML. “Is Age The Best Predictor For Non-Accidental Trauma (NAT) In Pediatric Patients With Femur Fractures?” ePoster Presentation at the Pediatric Orthopaedic Society Annual Meeting; Toronto, Canada; May 2013
    o Caplan, JL, Whitfield, B, and Neviaser RJ. Subscapularis Function After Primary Tendon to Tendon Repair in Patients After Replacement Arthroplasty of the Shoulder. Presented at 10th International Congress of Shoulder and Elbow Surgery; Salvador da Bahia, Brazil; September 2007
    o Cluett, J, Caplan, JL and Yu W. “Perioperative Cardiac Evaluation in Patients with Acute Hip Fractures. Poster at the American Academy of Orthopaedic Surgeons Annual Meeting; Washington, DC; February 2005
A. Personal Statement
I am Professor of Biology and Director of the Center for Integrative Genomics at Georgia Tech. My group conducts systems genomics research, building on 15 years of quantitative genetic research in Drosophila, but now with a focus on human genomics. We are primarily interested in the interaction between genotype and environment, the joint influences of these sources of variation on disease susceptibility, and the development of personalized disease risk profiles based on integration of genomic and clinical data. As a member of the Emory-GA Tech Predictive Health Initiative, we are pursuing genome-wide genetics of gene expression (GOGE) and genetics of metabolism studies of various cohorts in the Atlanta area. We are using next-generation sequencing approaches to study the regulation of gene expression, alternative splicing, and allele-specificity of transcription in the context of cardiac and metabolic disease, and in pediatric cohorts. I am part of an NIH Program Project on “Statistical and Quantitative Genetics” directed by Bruce Weir at the University of Washington, for which my role is development of new approaches to quantifying the contributions of rare and common variants to transcriptional regulation in peripheral blood. I also direct the functional genomics arm of the NIAID “Malaria Host Pathogen Interaction Center” contract investigating how non-human primates respond to Plasmodium infection. I will also direct a new NIGMS Training Grant in Computational Biology and Bioinformatics at Georgia Tech. My laboratory combines wet lab assays with statistical genetic and bioinformatics research. I serve as section editor for natural variation at PLOS Genetics, and am an elected member of the AAAS.

B. Positions and Honors

Positions and Employment
1990-1993 Post-Doctoral Fellow, Hogness group, Developmental Genetics, Stanford University, CA
1994-1994 Post-Doctoral Fellow, Laurie group, Evolutionary Genetics, Duke University, Durham, NC
1994-1998 Assistant Professor, Dept of Biology, University of Michigan, Ann Arbor, MI
1998-2004 Asst then Assoc Professor, Dept of Genetics, North Carolina State University, Raleigh NC
2005-2007 William Neal Reynolds Professor, Dept of Genetics, NC State University, Raleigh NC
2008-2009 Professor, School of Integrative Biology, University of Queensland, Brisbane, Australia
2009- Professor, School of Biology, Georgia Institute of Technology, Atlanta, GA

Other Experience and Professional Memberships
2013-2014 Consultant on systems genomics to NICHD, Biostatistics and Bioinformatics Branch
2004-2007 Assistant Director for Life Sciences, College of Ag and Life Sciences, NCSU, Raleigh NC
2005-2007 Associate Director, National Evolutionary Synthesis Center, Durham NC
2006-present Section Editor, Gene Expression and Natural Variation, PLoS Genetics
2007-present Chair, Advisory Board, Max Plank Institute for Evolutionary Biology, Ploen Germany
2003-2007 Member, FlyBase Advisory Board
2002-2005 Chair, North Carolina Governor’s Task Force on Genomics and Public Health
Honors
2010-2014 Tutor, Leena Peltonen School of Human Genetics, Hinxton UK
2008 Recipient of Australian Professorial Fellowship from Australian Research Council
2006 Elected Fellow of the American Association for the Advancement of Science.
2005 Named William Neal Reynolds Distinguished Professor of Genetics, NCSU
2002 NCSU Alumni Outstanding Researcher Award
1996-2001 David and Lucille Packard Foundation Fellow in Science and Engineering
1996-1997 Basil O’Connor Young Investigator of the March of Dimes Research Foundation
1990-1993 Helen Hay Whitney Foundation Post-Doctoral Fellow

C. 15 Selected recent peer-reviewed publications (last 4 years) (From 107 Papers & 2 Books)


**D. Research Support**

**Ongoing Research Support**

1 P01GM099568 (Gibson Project; BS Weir Program PI, Univ Washington) 05/01/12 - 04/30/17
National Institutes of Health, General Medical Sciences
Statistical Genetics Program Project, Project 3
Role: PI of experimental project to quantify rare and common variant influences on expression of 96 genes by deep sequencing of regulatory regions and Fluidigm-based qRT-PCR of over 3,000 people from healthy, cardiac, and Crohn’s disease cohorts. Other 4 Projects are purely statistical methodology development.

NIAID-DMID-NIHAI2010100 (M. Galinski, Yerkes Primate Center, PI) 09/15/12 - 09/14/17
MaHPIC: An Integrated approach to understanding host-pathogen interactions in malaria
Role: Co-PI for Georgia Tech component (Emory, UGA, CDC other partners)
Comparative RNA-Seq of five non-human primate-Plasmodium infection pairs throughout the course of infection and recovery, combined with systems modeling of the dynamic mechanisms of host and pathogen response.

1 T32 GM105490-01 (G Gibson, Georgia Tech, PI) 07/01/14 - 06/30/19
A Computational Biology and Predictive Health Genomics Training Program at GT
Role: PI for 20 Primary instructors
Verbal notification of award indicates 4 training slots will be awarded in Year 1.

**Completed Research Support (past 3 years)**

R01HL085481 (Co-PI) 01/01/08 - 12/30/12
National Institutes of Health, Heart, Lung and Blood
Polygenic Basis of Cardiac Dysfunction in *Drosophila*. 
Role: Gibson, R. Bodmer - Burnham Institute, San Diego, Co-PIs

Georgia Tech Institute for Bioengineering and Bioscience (IBB) 7/01/11 – 6/30/13
Single cell genomic profiling
Role: Co-PI with Melissa Kemp, GT
Fluidigm-based single cell qRT-PCR in the aging immune system to evaluate the prevalence of functional subclasses of neutrophils and T-cells and their interaction

Childrens Healthcare of Atlanta (CHOA) 06/01/13 - 05/31/14
Toward Personalized Treatment for Osteochonritis Dissecans based on Genomic and Micro-CT Imaging
Role: Co-PI with Bob Guldberg, GT
RNA-Seq analysis of bone biopsies to classify disease severity and etiology
BIOGRAPHICAL SKETCH

Provide the following information for the key personnel in the order listed for Form Page 2. Follow this format for each person. DO NOT EXCEED FOUR PAGES.

NAME
Robert C. Jerris

POSITION TITLE
Director, Clinical Microbiology
Children’s Healthcare of Atlanta

EDUCATION/TRAINING (Begin with baccalaureate or other initial professional education, such as nursing, and include postdoctoral training.)

<table>
<thead>
<tr>
<th>INSTITUTION AND LOCATION</th>
<th>DEGREE</th>
<th>YEAR(s)</th>
<th>FIELD OF STUDY</th>
</tr>
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<tbody>
<tr>
<td>Florida State University, Tallahassee, Florida</td>
<td>B.S.</td>
<td>1975</td>
<td>Microbiology and Medical Technology</td>
</tr>
<tr>
<td>Emory University, Atlanta, Georgia</td>
<td>M. Sc.</td>
<td>1978</td>
<td>Clinical Microbiology</td>
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<tr>
<td>Emory University, Atlanta, Georgia</td>
<td>Ph.D.</td>
<td>1981</td>
<td>Experimental Pathology</td>
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<tr>
<td>Postdoctoral Fellowship, Centers for Disease</td>
<td></td>
<td>1981-1983</td>
<td>Medical and Public Health Laboratory Microbiology</td>
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</table>

NOTE: The Biographical Sketch may not exceed four pages. Items A and B (together) may not exceed two of the four-page limit. Follow the formats and instructions on the attached sample.

1. **Positions and Honors.** List in chronological order previous positions, concluding with your present position. List any honors. Include present membership on any Federal Government public advisory committee.

**Employment / Experience**

1973-1975 Technologist, Division of Instructional Research Services, Florida State University, Tallahassee, Florida

1975-1976 Research Assistant, Emory University, Atlanta, Georgia

Research involved production of antiserum and evaluation of fluorescent antibody conjugates for rapid identification of anaerobic Bacteroides species

1981-1983 Postdoctoral Resident in Medical and Public Health Laboratory Microbiology, CDC, Atlanta, Georgia

1983-1984 Director, Microbiology Laboratories, Gwinnett Hospital System, Gwinnett County, Georgia

1984-1985 Laboratory, Clinical Program Director, Community Health Section Laboratory

Georgia Department of Human Resources Atlanta, Georgia

1983-1998 Consultant, Clinical Microbiology

1998-PRESENT Lecturer, visiting Professor

College of Health and Human Sciences

Georgia State University

Atlanta, GA

1993-1998 Assistant Professor, Adjunct

School of Medical Technology

Georgia State University

Atlanta, GA

1983-PRESENT Assistant Professor, Clinical

Emory University School of Medicine,
Department of Pathology and Laboratory Medicine
Atlanta, GA
1987-1997 Consultant, Anaerobic Bacteriology
Emory University Dental School (PERL)
Department of Oral Pathology
Atlanta, Georgia
1986-1992 Director, Clinical Laboratory Sciences Program
Emory University School of Medicine, Department of Pathology,
Atlanta, Georgia
1998-Present Microbiology Technical Resources
Clinical Microbiology Consulting
1985-4/2004 Director, Clinical Microbiology Laboratories
Department of Pathology
DeKalb Medical Center
Committees: Pharmacy and Therapeutics
Infection Control Education
2004-PRESENT Director, Clinical Microbiology
Children’s Healthcare of Atlanta (Henrietta Egleston Medical Center
and Scottish Rite Medical Center)

Honors
ASM; College of the American Academy of Microbiology; Board Member 2006-2009, 2009-
2012; Professional Affairs Committee 2012-2015.


Lead Scientist : Assessment of Laboratory Capacity in Rural Thailand for the Ministry of Public
Health (Thailand) and the International Emerging Infections Program- Thailand –
CDC, 10/03

Lead scientist: Establishing Blood cultures for Rural Guatemala-IEIP- 9/07

State of Georgia:
Clinical Microbiology representative and Alternate to the Clinical Laboratory, Blood
Bank, and Tissue Bank Advisory Council (1985-2005)

American Society for Microbiology:
1987-1996, Oral Examination Committee, American Board of Medical
Microbiologists (ABMM)-Development and Analysis of Board Questions
1989-1992, Standards and Examination Committee
1993-1996, Standards and Examination Committee
1992- Chairman, Nominations Committee, Division C.
1996- Chairman, Nominations Committee, Division C
2000-2003-Waksman Foundation Lecturer
2003- Division C- Ask the Experts
2003- Chairman, Nominations Committee Division C
Georgia State University, Outstanding Service Award, Dept. Med Techn. 1994
2. **Recent, Selected peer-reviewed publications (in chronological order).** Do not include publications submitted or in preparation.


Current Activity

Presentations/Publications:


Current Research:

- Matrix Assisted Laser Desorption Ionization Time of Flight for:
  a. Identification of routine clinical isolates
  b. Modification of MALDI-ToF techniques for identification of rapidly growing mycobacteris sp.
  c. Detection of carbapenemase activity for susceptibility testing

- Multiplex Viral PCR Testing for respiratory pathogens: impact on workflow, cost and outcomes.
• Collaboration CF protocol (The EPIC Observational Study: Longitudinal Assessment of Risk Factors For and Impact of *Pseudomonas aeruginosa* Acquisition and Early Anti-Pseudomonal Treatment in Children with CF)

• Collaboration CF protocol (A Multi-Center, Double Blind, Placebo Controlled Randomized, Efficacy and Safety of Denufosol Tetrasodium Inhalation Solution in Patients with Mild CF)

• Center for Disease Control and Prevention Collaborative Study with BRATT Lab to serve as SUBJECT MATTER EXPERT LAB in defining the specificity of biological reagents using RT-PCR for detection of agents of Bioterrorism

• Methicillin Resistant *Staphylococcus aureus* (MRSA) in the Emergency Department (ED): Relationship between Colonization and Skin and Soft Tissue Infections (SSTI) and Treatment Outcomes. LC Immergluck, R Jerris, S Jain
NAME  
Lars Frederick Westblade

POSITION TITLE  
Assistant Professor
Associate Director of Microbiology and Immunology

eRA COMMONS USER NAME (credential, e.g., agency login)  

EDUCATION/TRAINING (Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable.)

<table>
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<tr>
<th>INSTITUTION AND LOCATION</th>
<th>DEGREE (if applicable)</th>
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<th>FIELD OF STUDY</th>
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<tr>
<td>The University of Birmingham, UK</td>
<td>B.Sc (Hons)</td>
<td>09/95-06/98</td>
<td>Biochemistry with Biotechnology</td>
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<tr>
<td>The University of Birmingham, UK</td>
<td>M.Phil (Res)</td>
<td>09/98-09/99</td>
<td>Molecular and Cellular Biology</td>
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<tr>
<td>The University of Birmingham, UK</td>
<td>Ph.D</td>
<td>09/99-09/02</td>
<td>Biochemistry</td>
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<td>The Rockefeller University, USA</td>
<td>Postdoctoral</td>
<td>05/03-06/08</td>
<td>Molecular Biophysics/Biology</td>
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<tr>
<td>Washington University in St. Louis, USA</td>
<td>Fellowship</td>
<td>07/10-06/12</td>
<td>Medical and Public Health Microbiology</td>
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</table>
A. Personal Statement
The goal of the proposed study is to develop rapid, sensitive/specific methods to diagnose osteoarticular infections in children. We propose to analyze clinical bone and fluid specimens using a novel real-time polymerase chain reaction (PCR)/microfluidic platform. I have extensive experience with molecular biology techniques and microfluidic systems. This, coupled with my training as a clinical microbiologist (certified by the American Board of Medical Microbiology), ensures that I will be able to assist the study from specimen procurement, microbiological analysis to method development.

B. Positions and Honors

**Positions and Employment**

<table>
<thead>
<tr>
<th>Date</th>
<th>Position</th>
<th>Institution</th>
<th>Location</th>
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<tbody>
<tr>
<td>07/08–12/08</td>
<td>Scientist, Protein Engineering</td>
<td>Amyris Biotechnologies</td>
<td>Emeryville, CA</td>
</tr>
<tr>
<td>07/09–06/10</td>
<td>Asst. Prof., Dept. of Pharmaceutical/Biomedical Sciences</td>
<td>Touro College of Pharmacy</td>
<td>New York, NY</td>
</tr>
<tr>
<td>08/12–08/13</td>
<td>Dir. of Microbiology</td>
<td>Long Island Jewish Medical Center</td>
<td>New Hyde Park, NY, USA</td>
</tr>
<tr>
<td>08/13–Present</td>
<td>Assoc. Dir. of Clinical Microbiology and Immunology</td>
<td>Children’s Healthcare of Atlanta</td>
<td>Atlanta, GA, USA</td>
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**Honors**

<table>
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<th>Date</th>
<th>Honor Description</th>
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<tbody>
<tr>
<td>06/11</td>
<td>Paul E. Strandjord Young Investigator Award, Academy of Clinical Laboratory Physicians and Scientists</td>
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<tr>
<td>09/99–09/02</td>
<td>United Kingdom Biotechnological and Biological Sciences Research Council Graduate Fellowship</td>
</tr>
</tbody>
</table>

C. Selected Peer-reviewed Publications

*These authors contributed equally

#Corresponding author


NAME
Craig Andrew Shapiro

POSITION TITLE
Pediatric Infectious Disease Physician
Adjunct instructor in Pediatric Infectious Diseases

EDUCATION/TRAINING  
(Begin with baccalaureate or other initial professional education, such as nursing, and include postdoctoral training.)

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<td>Colgate University, Hamilton, NY</td>
<td>B.A.</td>
<td>1998-2002</td>
<td>Molecular Biology</td>
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<tr>
<td>SUNY Downstate College of Medicine, Brooklyn, NY</td>
<td>M.D.</td>
<td>2002-2006</td>
<td>Medicine</td>
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<tr>
<td>George Washington University School of Medicine, Washington, DC</td>
<td></td>
<td>2006-2009</td>
<td>Pediatric Residency</td>
</tr>
<tr>
<td>Emory University School of Medicine, Atlanta, GA</td>
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<td>2009-2012</td>
<td>Pediatric Infectious Diseases Fellowship</td>
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A. Personal Statement
I am a pediatric physician specializing in infectious diseases. I have experience in conducting clinical and translational research with a focus on diagnostic tools to better define disease states. As a pediatric infectious disease fellow I worked on a research project to elucidate the CD8 T-cell response of infants to Respiratory Syncytial Virus (RSV) using a novel Elispot assay. I was involved in all stages of this research, including writing up the proposal, enrollment of subjects and testing of samples in the laboratory. My experience in enrolling infant subjects into a clinical study will be of importance as we look to determine the effectiveness of a novel molecular assay to diagnose osteoarticular infections in children. I have also performed prospective reviews of novel laboratory based assays to determine the impact on clinical care. The current application will allow me to utilize the skills listed above, and I have chosen co-investigators (Drs. Anderson, Gibson, and Jadhao) who provide additional expertise in molecular diagnostics and the laboratory methods necessary to test the patient samples I will be collecting.

B. Positions and Honors.

Positions and Employment:
2006-2009: Pediatrics Residency, Children’s National Medical Center, Washington, DC
2009-2012: Pediatric Infectious Diseases Fellowship, Emory University School of Medicine, Atlanta, GA
2012-present: Physician, Pediatric Infectious Disease Associates PC

Professional Memberships:
2006-present American Academy of Pediatrics
2009-present Infectious Diseases Society of America
2009-present Pediatric Infectious Diseases Society
2009-present American Academy of Pediatrics Section on Infectious Diseases

Honors:
2000: Howard Hughes Biological Sciences Internship, National Institutes of Health
2006: Distinction in Research, SUNY Downstate College of Medicine
2007: **25th Hour Award**, Children’s National Medical Center
Honored as the intern who dedicated the most time to his profession

2009: **Honorable Mention**, 9th Annual Children’s National Medical Center Research Day

2011: **Pediatric Infectious Diseases Society of America Fellows Travel Grant**, 49th annual meeting of the IDSA;

C. **Selected peer-reviewed publications (in chronological order).**


D. **Research Support**

**Ongoing**

“**Impact of a rapid PBP2a assay on outcomes of pediatric patients with Staphylococcus aureus infections**”
Assess the impact of a newly implemented rapid Staphylococcus aureus methicillin resistance identification assay on outcomes of hospitalized pediatric patients with *S. aureus* infections at Children’s Healthcare of Atlanta
Role: Co-investigator

“**Time to optimal antimicrobial therapy following implementation of MALDI-TOF MS and antimicrobial stewardship team interventions at an academic pediatric institution**”
Retrospective/prospective cohort study of clinical outcomes in patients with BSIs comparing MALDI-TOF MS analysis with antimicrobial stewardship intervention to conventional laboratory methods for identification of organisms from blood cultures. Role: Co-investigator

**Completed**

“**Identification of novel CD8+ T-cell epitopes in Respiratory Syncytial Virus as targets for vaccine design**”
Evaluated CD8+ T cell response in acutely infected infants with respiratory syncytial virus. Stimulated T cells with different subgroups of RSV to identify immunodominant epitopes for use as a potential vaccine target.
Role: Co-investigator
1. Introduction

Two out of every 5000 children under the age of 13 years are diagnosed with acute infectious osteoarthritis/septic arthritis. In one of the studies in United States published from University of Tennessee Health Science Center, Memphis, TN, the incidence of acute osteoarticular infection increased from 2.6 to 6 per 1000 admission between years 2004 to 2006. Children 6 months to 5 years of age are especially susceptible to the invasive bacterial pathogens associated with osteoarthritis (Yagupsky et al, 2011). Children younger than 6 month of age are presumably protected by maternally transferred antibodies to outer bacterial membrane protein antigens. Though older children, young adults, and adults are less at risk, (Ogawa et al, 2014, Yagupsky et al, 2011), they also can develop osteoarticular infections. The affected children usually present with fever, painful and swollen joints or bones and require prompt diagnostic evaluation and treatment (Yagupsky et al, 2011, Arnold et al, 2006).

Bacterial pathogens such as *Staphylococcus aureus*, *Streptococcus pyogenes*, *Streptococcus pneumoniae* are most often detected in these infections through culture in clinical microbiological laboratories. Unfortunately, epidemiologic studies show that the causative organism is detected in only 50-60% of cases, leaving up to 50% of cases with no diagnosis, when tissue, synovial fluid, and blood are obtained and cultured. Culture negative specimens can result from antibiotic administration before specimens are obtained, presence of difficult to culture, fastidious bacteria, improper specimen handling, or presence of novel pathogens. In recent studies using PCR, *Kingella kingae*, a fastidious, a gram negative coccobacillus was detected in up to 45% of cases of bone and joint infections in preschool children. *Kingella kingae* is increasingly recognized as a common cause of bone and joint infections in young children. Consequently, sensitive molecular detection assays, e.g. real time PCR, can improve our ability to detect bacteria in bone and joint infections. Another important challenge in the management of these infections is increase in antibiotic resistant organisms. Arnold et al noted an increase in methicillin-resistant *Staphylococcus aureus* (MRSA) from 4% to 40% between 2004 and 2006 (Arnold et al, 2006). Since many of the genes associated with antibiotic resistance have been identified, real PCR assays can be used to identify an organism's likely resistance to certain antibiotics. Recent molecular characterization of previously isolated *Kingella kingae* strains, in Minnesota during 2004-2007 in the United States, revealed presence of β-lactamase gene containing plasmid (Banerjee et al, 2013 and Basmaci et al, 2014). As noted previously *Kingella kingae* bacteria are difficult to culture. Detection of resistance to β-lactam group of antibiotics (Penicillin and its derivatives) which are preferentially used in treatment of osteoarticular infections and infective endocarditis prevention in high risk patients further underlines the significance for need of rapid molecular diagnostic assays that detect antibiotic resistance and bacterial pathogen simultaneously.

Thus, molecular testing should improve a clinician’s ability to manage bone and joint infections by increasing the frequency with which the bacterial pathogen is detected and quickly identifying likely antibiotic resistant organisms. To assess the potential for molecular testing to improve management of bone and joint infections in children treated at CHOA, we completed a small pilot study of joint infections in children seen at CHOA at Scottish Rite. In this study, 27 specimens were tested by PCR and culture and 13 of these grew *S. aureus*, 10 were culture
negative and 1 each grew a candida species, *S. epidermidus*, *S. pneumoniae*, and vancomycin-resistant enterococci (VRE). Ten of the *S. aureus* culture positive specimens were *S. aureus* PCR positive and 3 of the 10 culture negative specimens were PCR positive for *Kingella kingae*. We are now testing for antibiotic resistance genes and will compare these results with those from culture-based antibiotic resistance testing. These results show that the addition of molecular detection will significantly increase the number of specimens positive over culture testing alone.

We now propose to apply previously validated pathogen specific real time PCR assays to detect the bacteria, and add PCR testing for bacterial antibiotic resistance genes in clinical specimens from osteoarticular infections of children. To make testing multiple (9) real time PCR assays practical in a clinical setting, investigators at Georgia Tech will adapt the validated real time PCR assays to the Fluidigm microfluidic PCR system. The Fluidigm system automates testing up to 96 specimens against 96 real time PCR assays simultaneously and quickly. With the Fluidigm system, we can detect bacteria and resistance genes in <24 hours where culture detection usually requires 2-7 days and an additional 2-3 days is required to complete antibiotic resistance testing.

Drs. Flannagan and Shapiro will enroll patients, collect the relevant clinical, epidemiologic and laboratory data, and specimens for testing. Dr. Jadhao in Dr. Anderson’s laboratory at the Division of Pediatric Infectious Diseases, Emory University School of Medicine and CHOA, will complete development and validation of real time PCR assays for antibiotic resistance genes and primers and probes for bacterial pathogen PCR assays (*Jadhao et al*, 2014) and test specimens by individual PCR assays. The interpretation of results will be done in consultation with Drs. Jerris and Westblade. Dr. Gibson’s laboratory at Georgia Tech will adapt these assays to the Fluidigm system and test specimens with the Fluidigm system. Dr. Shapiro will lead analysis of cost and management benefits of adding molecular testing to culture testing for diagnosis of osteoarticular infections.

2. Questions to be addressed by the Proposed Research with Specific Aims

**Aim 1.** Determine the increase in rate and speed of detecting bacterial pathogens and antibiotic resistance in osteoarthritis infections with real time PCR testing as an adjunct to culture detection and characterization of pathogens.

**Aim 2.** Determine the sensitivity and specificity of the Fluidigm system for detecting bacteria and antibiotic resistance in osteoarthritis infections compared to culture-based and individual real time PCR detection systems.

**Aim 3.** Estimate the potential decrease in healthcare costs and increase in patient safety and treatment efficacy when molecular diagnostic assays are combined with culture to diagnose osteoarticular infections.

The resulting data will determine sensitivity of PCR for detecting bacteria in culture positive specimens, the frequency with which PCR detects bacteria in culture negative specimens, and the frequency with which resistance genes are detected and predict resistance in comparison to standard culture techniques. These data will then be used to determine the potential cost,
patient safety, and increased treatment efficacy with PCR testing. Currently, the clinical
diagnosis of osteoarthritis/septic arthritis in children is based on clinical presentation (fever,
painful and swollen joint, difficulty/inability to bear weight and laboratory tests results (mainly
WBC count, ESR, CRP) and X-ray/MRI imaging of affected joints. The culture based
identification is current gold standard for detecting the etiologic agent. However, it has high
false negative result rates. The empirical antibiotic treatment of affected children without
confirmatory diagnosis of bacterial pathogen compromises optimal clinical management and
prognosis. We propose to develop and implement novel molecular diagnostic assays that can
detect both specific pathogen and antibiotic resistance gene based on real time PCR and a
microfluidics based (Fluidigm) automated real time PCR system. Real time PCR diagnostics
should both 1) improve our ability to detect pathogens and 2) reduce time to detection from
several days to a few hours. Thus, molecular diagnostics should help the clinician focus
antibiotic therapy quickly and decrease costs and the risk of adverse outcomes associated with
broad spectrum antibiotic therapy while ensuring therapy is appropriate to the infecting
organism. The proposed study will document the potential for molecular diagnostics to improve
management of children with osteomyelitis and septic arthritis.

3. Proposed Study Methodology Including Data Sources

Children >6 months to 18 years of age with signs and symptoms of septic arthritis or
osteomyelitis, i.e. fever, painful and/or swelling over bones or at joints, radiologic evidence of
bone or joint infection, will be recruited by the investigator or research coordinators from
patients admitted to Emergency room at Egleston and Scottish Rite campuses. The study
coordinator and/or investigator will secure informed consent and assent as indicated and,
enrolled, the investigators will secure aliquots of specimens collected for microbiologic studies,
e.g. aspirate or surgical specimen from the affected joint or bone for PCR testing. All procedures
will be done as part of the standard work up and care of the child. Some specimens will be
collected in the emergency room, e.g. aspiration of infected joints, while other specimens will be
collected during surgery, e.g. to diagnose and/or drain infected bones. Specimens for the PCR
testing will be aliquots of specimens collected aseptically for culture studies that will be done in
CHOA clinical microbiology laboratories. The sample aliquot will be identified by a number
linked to the patient and no clinical data, epidemiologic data, or personal identifiers will
accompany the specimens. The specimens will be stored at -80°C until testing. The investigator
or study coordinator will collect clinical, laboratory, and radiologic data from the patient’s
medical record. For testing, total DNA will be extracted from the specimen using the Kingfisher
automatic extractor. One aliquot of extracted DNA will be tested at the laboratory in Division of
Pediatric Infectious Diseases by individual real time PCR assays. A second aliquot will be
transported to the Georgia Tech laboratory for testing using the Fluidigm system. Specimens
will be tested by real time PCR for 5 bacterial pathogens, S. aureus, S. pyogenes, S.
pneumoniae, K. Kingae, and Group B Streptococcus (GBS), and 4 antibiotic resistance genes,
beta-lactamase, mecA, mecC and ermA. The specimens will be batch tested. The patients
laboratory test results will be combined with the PCR study results to determine: 1) ability of a)
individual PCR and b) Fluidigm assays to detect the bacteria found in culture positive
specimens; 2) ability of a) individual PCR and b) Fluidigm assays to detect bacteria in culture
negative specimens; and 3) the ability of individual PCR assays detect bacteria positive by the Fluidigm assays and visa versa.

**Study Timeline**

<table>
<thead>
<tr>
<th>Study Timeline</th>
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</tr>
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<tbody>
<tr>
<td></td>
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<tr>
<td>Renew IRB protocol and secure approval to add osteomyelitis patients</td>
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<tr>
<td>Enroll patients and collect specimens</td>
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<tr>
<td>Develop and validate resistance gene assays</td>
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<td>Adapt bacterial PCR and resistance gene assays to the Fluidigm system</td>
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<tr>
<td>Test specimens by individual real time PCR assays</td>
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<tr>
<td>Test specimens with the Fluidigm real time PCR system</td>
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<tr>
<td>Analyze data and prepare manuscripts</td>
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**Table 2. Study tasks**

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<th>Study Tasks</th>
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<th>Post-Procedure</th>
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<td>Medical record review</td>
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<td>Individual real time PCR testing</td>
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<tr>
<td>Fluidigm real time PCR testing</td>
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</tr>
</tbody>
</table>

**4. Anticipated Results of the Study**

The main benefit of the proposed study will determining the value of adding molecular diagnostics to culture studies for the identification and likely antibiotic resistance of bacteria causing septic arthritis and osteomyelitis. Defining the value of molecular diagnostics in this setting will guide development and application of molecular testing in the clinical microbiology laboratory. The addition of molecular testing likely will improve the safety and efficacy of treating osteomyelitis and septic arthritis in children.

The study will also provide data on two different testing strategies, individual real time PCR and the automated microfluidic based Fluidigm real time PCR system and the frequency with which commonly detected pathogens are in children with osteoarticular infections seen at CHOA.

**5. Budget with uses, justifications, and any other source of support or available funds**

(See attached NIH 398 Budget Form)

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<th>Category</th>
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<th>Year 2</th>
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**Budget Justification**
Personnel: Samad Jadhoa (3.0 months) will develop and validate real time PCR assays, process study specimens, extract DNA for real time PCR testing, do individual real time PCR testing, provide DNA for Fluidigm testing, manage laboratory data and participate in writing papers for publication.

Supplies: Miscellaneous supplies for testing and specimen storage, nucleic acid extract and PCR kits, and PCR primers and probes.

Other expenses: Processing and testing specimens using the Fluidigm PCR system and supplies and shipping costs for specimen collection and transport.

References


## Detailed Budget for Initial Budget Period

**Direct Costs Only**

List **Personnel** (Applicant organization only)

Use Cal, Acad, or Summer to Enter Months Devoted to Project

Enter Dollar Amounts Requested (omitting cents) for Salary Requested and Fringe Benefits

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**Subtotals**

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**Consultant Costs**

|                      |                |             |              |              | 0              |                |               |

**Equipment** *(Itemize)*

|                      |                |             |              |              | 0              |                |               |

**Supplies** *(Itemize by category)*

- PCR primers and probes, DNA extraction reagents, PCR reagents, tubes, gloves, and PCR plates:

|                      |                | 3,503       |              |              |                |                |               |

**Travel**

|                      |                | 3,503       |              |              |                |                |               |

**Inpatient Care Costs**

**Outpatient Care Costs**

**Alterations and Renovations** *(Itemize by category)*

**Other Expenses** *(Itemize by category)*

- Specimen collection and shipping costs $2000
- Fluidigm testing - 50 specimens/year $3000

|                      |                | 5,000       |              |              |                |                |               |

**Consortium/Contractual Costs**

**Direct Costs**

|                      |                | 25,000      |              |              |                |                |               |

**Subtotal Direct Costs for Initial Budget Period** *(Item 7a, Face Page)*

**Consolidated/Contractual Costs**

**Facilities and Administrative Costs**

|                      |                | 25000       |              |              |                |                |               |

**Total Direct Costs for Initial Budget Period**

**PHS 398 (Rev. 08/12 Approved Through 8/31/2015) OMB No. 0925-0001 Form Page 4**
Program Director/Principal Investigator (Last, First, Middle):  Anderson, Larry J.

### BUDGET FOR ENTIRE PROPOSED PROJECT PERIOD

**DIRECT COSTS ONLY**

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<tr>
<th>BUDGET CATEGORY TOTALS</th>
<th>INITIAL BUDGET PERIOD (from Form Page 4)</th>
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**TOTAL DIRECT COSTS FOR ENTIRE PROPOSED PROJECT PERIOD**

$50,000

**JUSTIFICATION.** Follow the budget justification instructions exactly. Use continuation pages as needed.

**Personnel:** Samad Jadhoa (3.0 months) will develop and validate real time PCR assays, process study specimens, extract DNA for real time PCR testing, do individual real time PCR testing, provide DNA for Fluidigm testing, manage laboratory data and participate in writing papers for publication.

**Supplies:** Miscellaneous supplies for testing and specimen storage, nucleic acid extract and PCR kits, and PCR primers and probes.

**Other expenses:** Processing and testing specimens using the Fluidigm PCR system and supplies and shipping costs for specimen collection and transport.
PROTECTION OF HUMAN SUBJECTS

RISK TO HUMAN SUBJECTS

Justification for the involvement of human subjects. Osteoarticular infections of bones and joints caused by pathogenic bacteria are considered as pediatric emergency and pose significant challenge to mobility of young children. Rapid identification of the causative bacterial pathogen is important to provide optimal treatment and minimize the risk complications rendered by ineffective therapy. For example, without causative organism identification, the clinician is forced to use broad-spectrum antibiotics which increase the risks for side effects including clostridium difficile colitis. Epidemiologic studies show that the causative organism is detected in 50-60% of cases when tissue, synovial fluid, and blood are obtained and cultured. Thus, no organism is identified in up to 50% of cases when specimens are available. With culture the most commonly detected bacteria of osteoarticular infections in children are Staphylococcus aureus, Streptococcus pyogenes, and Streptococcus pneumoniae. Less is known about fastidious organisms often missed by culture. With molecular detection, we are finding other bacteria such as Kingella kingae, a gram negative coccobacillus, in osteoarticular infections. With PCR, Kingella kingae has been detected in up to 45% of cases of bone and joint infections in preschool children. The use of a rapid and sensitive PCR assays to detect Kingella kingae, other fastidious bacteria, and other difficult to culture specimens, e.g. from patients with prior antibiotic treatment, should improve our ability to detect the cause and manage osteoarticular infections in children. Another challenge to managing these infections is antibiotic resistance. Since bacterial genes associated with antimicrobial resistance have been identified, PCR assays can detect their presence and guide choice of antibiotic therapy. We propose to use pathogen-specific and gene-specific real time PCR assays to detect the bacteria and antibiotic resistance genes in specimens from children with osteoarticular infections. The resulting data will determine frequency that PCR diagnostics detect bacteria otherwise missed and improve antibiotic therapy. These data will be used to determine the potential cost savings, patient safety benefits, and increase in treatment efficacy that PCR testing can provide. Finally, since testing for common bacterial and resistance genes will require multiple different PCR assays, we will adapt the individual assays to the Fluidigm microfluidic PCR system. This system provides an easy and efficient way to run multiple PCR assays in parallel simultaneously. We will compare the sensitivity and specificity of this system to individual real time PCR assays run in parallel. These improvements in confirmatory diagnosis of osteoarthritis/septic arthritis will help attending physicians in providing specific antibiotics and help in optimal clinical management of patients.

Description of proposed human subject involvement. Osteoarticular infections of bones and joints caused by bacterial pathogen affect normal activities and pose a significant risk of permanent damage. Rapid identification of the causative bacterial pathogen is important to administer effective treatment and prevent complications. The proposed studies are designed to develop methodology for confirmatory laboratory diagnosis. The proposed study population is children belonging to age group >6 months to 18 years. Specimens for the study will be an aliquot from specimens otherwise collected for standard workup and management of the patient. The study will be under an existing IRB-approved protocol.

Sources of Materials.

Study children (>6 months to 18 years of age) will be recruited by the principal investigator or research coordinators from patients admitted to Emergency room at Egleston and Scottish Rite campuses. Children with a clinical diagnosis of osteoarticular infection, e.g. fever, painful and/or swelling over bones or at joints, radiologic evidence of bone or joint infection will be identified by the attending physician who will then notify the investigator of the suspected osteoarticular involvement. The study coordinator and/or investigator will secure informed consent and assent as indicated and, enrolled, the investigators will secure an aliquot of specimens collected for microbiologic studies, e.g. aspirate or surgical specimen from the affected joint or bone for PCR testing. The specimens will be an aliquot from specimens collected for standard work up and management of the patient. Each patient will be given a specific number to match PCR result. Medical data that will be collected for research purposes include: (1) Data abstracted from the medical record on demographic characteristics, medical history related to the bone or joint infection, and laboratory and radiologic results related to the infection. Confidentiality will be protected by using a unique study identifier for data and specimens. This unique study number, and no personal identifiers, will be used to label all specimens sent to
laboratory. Hard copies of materials with personal identifiers will be kept in locked file cabinets and secured offices. All electronic study records used for analysis will have unique study identifier, no personal identifiers, and be password protected. All reporting of data for publication purposes will use summary data and will exclude personal identifiers.

Potential Risks: The potential risks to the patients are minimal. No specimens will be collected specifically for the study. The synovial fluid and bone specimens will be collected as part of the standard care and management of children with bone and joint infections.

ADEQUACY OF PROTECTION AGAINST RISK
Recruitment and informed consent. The subjects (children) for the osteoarthritis/septic arthritis study will be recruited from children admitted to Emergency room at Egleston and Scottish Rite campuses. Patients eligible for the study will be ascertained by 1) review of children presenting for history of fever, sore throat, acute pain in joints/osteoarthritis/septic arthritis treatment to the Egleston and Scottish Rite hospital. The child’s parent or guardian will be contacted by the study coordinator, informed about the study, and asked about the child’s medical history. If the child meets entry criteria and the parent or guardian is interested in having their child participate, they will be consented and the child entered into the study. The collection of bone and joint specimens will be done as part of the normal clinical management of the patient and not for the study. Child’s parent or guardian will be consented and the child assented as indicate. Copies of the signed forms will be provided to the parent or guardian or mother and the original signed copies will become a part of the subject’s medical record.

Protections Against Risk
To limit study specific risks, no synovial fluid or bone specimens will be collected from children who are not otherwise having specimens collected. Only, personnel trained in collection of specimen collection will collect the specimens. Confidentiality will be protected by substituting a unique study identifier for personal identifiers. This unique study number will be used to label all specimens and all data that leaves the local study site. The unique study number will be the only identifier associated with specimen during testing and data analysis related to the study. Hard copies of materials with subject identifiers will be kept in locked file cabinets and secured offices at the study site. All electronic records will be password protected.

POTENTIAL BENEFITS OF THE PROPOSED RESEARCH
There are no direct benefits to the study subjects from participating in this study. Specimens will be batched and tested later and results will be used only for study purposes and not for clinical management of patients. The potential benefits lie in the translation of knowledge from this study to improve our ability to treat infectious osteoarthritis/septic arthritis. The risks to the subjects are minimal and the potential gain in knowledge for future patients is substantial.

IMPORTANCE OF THE KNOWLEDGE TO BE GAINED
As one of the most important causes of serious infections in the young children, infectious osteoarthritis/septic arthritis is a high priority for confirmatory laboratory diagnosis. Thus, data generated by this project which should assist physicians in providing optimal therapy and clinical management and will be of substantial health benefit to young children in the future.
DATA AND SAFETY MONITORING PLAN
Since no treatment or intervention is included in the study and the study procedures are of minimal risk a Data and Safety Monitoring Plan has not been developed.

WOMEN AND MINORITY INCLUSION.
The study will only include children in age group of >6 months to 18 years. We will not exclude children based on gender or ethnicity. All eligible children who meet entry criteria and whose parents consent will be included.

INCLUSION OF CHILDREN
Children in age group of >6 months to 18 years are target population for this study.