SUMMARY STATEMENT

PROGRAM CONTACT: [redacted] **(Privileged Communication)** ***Release Date:*** [redacted] **11/17/2016**

Application Number: **1 K01 AI125413-01A1**

Principal Investigator

AMBROGGIO, LILLIAM

Applicant Organization: CINCINNATI CHILDRENS HOSP MED CTR

Review Group:	**MID-B** **Microbiology and Infectious Diseases B Subcommittee** **MID-B November 2016**
Meeting Date:	**11/04/2016**
Council:	**JAN 2017**
Requested Start:	**04/01/2017**
RFA/PA:	**PA16-190**
PCC:	**M59**

Project Title:	**Metabolomics Evaluation of the Etiology of Pneumonia**
SRG Action:	**Impact Score:** [redacted]
Next Steps:	**Visit http://grants.nih.gov/grants/next_steps.htm**
Human Subjects:	**30-Human subjects involved - Certified, no SRG concerns**
Animal Subjects:	**10-No live vertebrate animals involved for competing appl.**
Gender:	**1A-Both genders, scientifically acceptable**
Minority:	**1A-Minorities and non-minorities, scientifically acceptable**
Children:	**2A-Only Children, scientifically acceptable**
	Clinical Research - not NIH-defined Phase III Trial

Project Year	**Direct Costs Requested**	**Estimated Total Cost**
1		
2		
3		
4		
5		
TOTAL	[redacted]	[redacted]

ADMINISTRATIVE BUDGET NOTE: The budget shown is the requested budget and has not been adjusted to reflect any recommendations made by reviewers. If an award is planned, the costs will be calculated by Institute grants management staff based on the recommendations outlined below in the COMMITTEE BUDGET RECOMMENDATIONS section.

1K01AI125413-01A1 Ambroggio, L

AUTHENTICATION OF KEY BIOLOGICAL AND/OR CHEMICAL RESOURCES: UNACCEPTABLE

RESUME AND SUMMARY OF DISCUSSION: This exceptional resubmission application for a Mentored Research Scientist Development Award (K01) entitled Metabolomics Evaluation of the Etiology of Pneumonia was submitted by Lilliam Ambroggio, PhD, of Cincinnati Childrens Hospital Medical Center.

This is a resubmission of a K01 application from Lilliam Ambroggio, PhD MPH who is a trained epidemiologist and an Assistant Professor of Pediatrics at the University of Cincinnati Children's Hospital. She has an interest in translational research involving childhood diseases and metabolomics – the study of metabolites arising from the host-pathogen interaction that can provide rapid diagnostic information to improve therapy and outcomes. Her specific interest is in community-acquired pneumonia (CAP) in young children too young to produce a sputum sample, where diagnostics to confirm a pathogen is particularly difficult. She has expanded from two to three aims in this submission to address prior comments. Aims include: 1) to characterize the impact of age and gender on a healthy child's metabolome, 2) to compare urine metabolite profiles in non-infected children vs children with confirmed viral vs typical bacterial vs atypical bacterial CAP, and 3) to determine the impact of antibiotic therapy on serial sampling of urine metabolites in children with and without CAP.

Among strengths of this exceptional application is the candidate who has been highly productive throughout her career and has already had a major impact in her field. The project (a robust clinical study with comprehensive sample and data collection in young children presenting to the hospital with CAP) is one of great importance and the candidate supports its feasibility with substantial preliminary data. The panel agreed that she has had strong mentoring and institutional support in the past and this will continue based on the application's letters of support. The candidate has addressed all the concerns of the previous review including reducing her didactic load and restructuring her research plan. The panel was unanimous in judging this application as having exceptional impact in clinical diagnostics.

The only minor issue was whether the applicant was too advanced for a K award. The panel came to the conclusion that the candidate does require further training in the area of NMR metabolomics and bioinformatics in order to achieve her career goals. Furthermore she presents a timeline for submission of an R01. The panel's enthusiasm was not diminished by this issue. Based upon the evaluation of scientific and technical merit, this application received an Overall Impact/Priority score of [redacted].

DESCRIPTION (provided by applicant):
Lilliam Ambroggio, PhD, MPH is an infectious diseases epidemiologist whose overarching career goal is to improve outcomes for children with common, serious infections by developing methods to improve diagnostic accuracy and implementing these methods into clinical practice. The research she proposes entitled Metabolomic Evaluation in the Etiology of Pneumonia (MEEP) combines advanced statistical techniques with 1H-Nuclear Magnetic Resonance (NMR) metabolomics methodology to identify metabolites, which will facilitate etiologic diagnosis of community-acquired pneumonia (CAP) in children. Such pathogen identification will result in timely and accurate diagnosis that will permit targeted and effective management of this disease. Candidate: Dr. Ambroggio is an Assistant Professor of Pediatrics with a joint appointment in the Divisions of Hospital Medicine and Biostatistics and Epidemiology at Cincinnati Children's Hospital Medical Center (CCHMC). She completed a Master's in Public Health and a Ph.D. in Epidemiology at Drexel University prior to beginning a post-doctoral

fellowship at CCHMC. Her previous work in the clinical management of CAP focusing on antibiotic prescribing and diagnostic tools to detect pneumonia in combination with her previous training in molecular and cellular biology have prepared her to conduct the proposed research. The proposed career development plan will build upon her previous training with four training goals to enhance her trajectory toward becoming an independent investigator: 1) Experiential and didactic learning in study design and execution of quantitative 1H-NMR metabolomics; 2) Acquire and apply advanced statistical analyses; 3) Interpret metabolomics data and its biological context and 4) Develop leadership and professional skills to execute multicenter studies. Dr. Ambroggio proposes training activities that include didactic and experiential learning to enable her to gain the necessary skills for metabolomic research. Mentors/Environment: Dr. Ambroggio and her primary mentor, Samir S. Shah, MD, MSCE, have assembled a strong team of co-mentors and advisors to guide Dr. Ambroggio through the proposed training and research activities. The proposed career development plan utilizes the intellectual and metabolomics resources available through the University of Cincinnati and CCHMC, as well as resources available at the University of Michigan through Dr. Ambroggio's external mentor, Kathleen Stringer, PharmD. In addition Dr. Ambroggio will attend national seminars and workshops when optimal training is not available locally. As an institution CCHMC is committed to supporting junior faculty members through internal grants, administrative support and structured opportunities for faculty networking and education. Dr. Ambroggio will be obtaining biological specimens for this proposal from a fully operational, externally-funded prospective cohort study, CARPE DIEM. Both the ED and inpatient services at CCHMC provide an established research infrastructure and a large ambulatory and hospitalized patient population to conduct the proposed research. In addition all mentors have agreed to participate on Dr. Ambroggio's scholarly oversight committee which has been meeting quarterly since 2013. Research: There is currently no accurate method to identify the etiology of CAP in children. This results in overtreatment with antibiotics or delays in appropriate treatment in children who are at risk for CAP-related morbidity. This proposal is the first step in developing a specific, fast and noninvasive approach for pathogen identification in children diagnosed with CAP. Aim 1 characterizes the sources (e.g. age and sex) of variation that exist in a healthy child's metabolome over three points. Aim 2 compares metabolite profiles from children who had a positive PCR test from either the nasopharynx or the blood for a virus, bacterial infection such as Mycoplasma pneumoniae, or Streptococcus pneumoniae with children who have no known infections. The purpose of this aim is to identify unique metabolite profiles using quantitative 1H-NMR for each pathogen identified. Aim 3 will use urine samples from patients with CAP and compare them with samples from healthy contorls over three time points to determine the impact of antibiotic treatment on metabolite profiles. The completion of these aims will generate a metabolite profile database of common pathogens associated with childhood CAP and drive a systems biology approach to CAP diagnosis and treatment. Summary: The innovation of the proposed research is the integration of robust clinical phenotype data from an ambulatory population with quantitative NMR metabolomics using novel statistical methods to address the clinically challenging problem of pediatric CAP diagnostics. The strong collaborations between the Divisions of Hospital Medicine, Biostatistics and Epidemiology, and Emergency Medicine at CCHMC and with the metabolomics core at the University of Michigan ensure the success of the proposed research. This award will provide Dr. Ambroggio with the training and research needed to be successful in a future, multi-center study to validate the metabolite profiles of pathogens causing CAP in children. Furthermore, this career development award will facilitate Dr. Ambroggio's development into a nationally-recognized independent investigator and leader conducting research that improves diagnostic tools for children with infectious diseases, specifically CAP.

PUBLIC HEALTH RELEVANCE

Project Narrative Community-acquired pneumonia (CAP) causes substantial morbidity in children. It is the fifth most common cause of pediatric hospitalization and, cumulatively, the most costly among infants and children. Currently there is no single test available that can distinguish between the >10

pathogens that cause CAP. The objective of this prospective cohort study is to determine distinct metabolite profile for major classes of pathogens that cause CAP in children (e.g. virus, typical and atypical bacteria). Urine and blood samples will be collected from children diagnosed with CAP in the Emergency Department or who are hospitalized at Cincinnati Children's Hospital Medical Center. Urine from children whose PCR tests are positive for a virus, typical bacteria or atypical bacteria will be sex and aged-matched to control samples. All urine samples will then be evaluated using quantitative 1H-nuclear magnetic resonance. Advanced statistical methods will be applied to the identified metabolite dataset to determine unique metabolite profiles. These profiles will be used as the foundation for future studies in the diagnostic capabilities of 1H-NMR for pathogen identification in children diagnosed with CAP. In addition, the concepts learned through this grant, as well as the career development pursued by the investigator, will be readily applicable to diagnostic testing for multiple infectious diseases.

CRITIQUE: The comments in the CRITIQUE section were prepared by the reviewers assigned to this application and are provided without significant modification or editing by staff. They are included to indicate the range of comments made during the discussion, and may not reflect the final outcome. The RESUME AND SUMMARY OF DISCUSSION section summarizes the final opinion of the committee after the discussion and is the basis for the assigned Impact/Priority score.

Critique 1

Candidate:	1
Career Development Plan/Career Goals & Objectives:	1
Research Plan:	1
Mentor/Co-Mentor(s), Consultant(s), and Collaborator(s):	1
Environment and Institutional Commitment to the Candidate:	1

Overall Impact:
This is a resubmission from a stellar candidate who has already achieved a focused and successful early career as an ID epidemiologist in pediatric CAP. She has outstanding institutional support and has created and received multi-year internal funding for an extensive infrastructure to consent and sample children in emergency departments who present with CAP. Specifically, this study is embedded in a robust clinical study with comprehensive sample and data collection in young children presenting to the hospital with CAP. She has achieved important and compelling preliminary data to further her study on detecting the etiology of CAP (viral, atypical bacterial, or typical bacterial) using metabolomics, which has the potential to greatly impact clinical care and antibiotic stewardship. Her career development plan is solid. She has outstanding mentorship across all her mentors. She has addressed all concerns in a focused and comprehensive manner. The aims are all well designed and conceived. High impact application.

1. Candidate:
Strengths

- Longstanding dedication to research. Research tech for 3y at Fred Hutchinson led to pursuit of MPH and PhD in Epidemiology at Drexel (2011). Completed post-doc at U Cincinnati and was promoted to Assistant Professor of Pediatrics in 2013, joint appointment in Hospital Medicine and Biostatistics and Epidemiology at Cincinnati Children's Hospital Medical Center
- Has prior research foundation for the study of CAP. 9 of 14 papers are first-author papers; another first author paper in press. 4 are reviews/commentaries. Prior publications include papers in Pediatrics and PIDJ, and are highly relevant to CAP, including antibiotic selection, comparative effectiveness of treatment regimens, determinants of illness severity, and biomarkers for CAP diagnosis and severity.
- Successful young investigator, 5y out from her PhD. Had received two prior external non-federal grants - a 1y society award (ended 2014) (American Pediatric Association) for impact of pediatric CAP guidelines and a Thrasher fund award (ended 2014) for evaluating chest ultrasound on diagnosis of CAP. Recently finished a 3y institution-specific research grant on CAP and metabolomics that ended Dec 2015 and provided prelim data for this grant (CARPE DIEM). Institutional research funds have now been continued till Dec 2018 and supports the infrastructure for the sample collections for this K.
- She is a co-PI on other active pediatric pneumonia grants through end-2017. Given end dates, should be able to secure focused effort on this K.
- Strong letters of recommendation
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Weaknesses

- Potentially too advanced in her career success for a K, but this is offset by the skillset, particularly in NMR metabolomics that she wishes to achieve. Also notable that her plans for her first R01 are to pursue a large multi-center validation study.

2. Career Development Plan/ Career Goals & Objectives:

Strengths

- Interested in translational science between childhood disease and metabolomics, with a focus on epi-metabolomics, the combination of large-scale population health assessments with metabolomics. Long term goal is to identify a metabolite predictive of CAP in children and to validate it in multi-center studies.
- Has target skill sets for career development – expertise in use and analysis of NMR metabolomics, improved interpretation of metabolomics data, advanced statistics, and multi-center study design/implementation
- Longstanding mentoring committee with quarterly meetings since 2013. Will continue weekly meetings with Dr. Shah as her primary mentor, and every other week calls with Dr. Stringer, her mentor for metabolomics. She will have monthly meetings with her co-mentor on molecular epi, and monthly meetings with Shah/Ruddy/Florin in development of a pediatric cohort on which her prelim data are based.
- Has addressed the concern for an ambitious training schedule, previously planned as 6 courses over 2 years. She has since completed 2 of the courses and now proposes 3 formal classes over 3 years (structural equation modeling, medical microbio, and ethics in

research) to complement her hands on training in NMR metabolomics and molecular epidemiology.

- Well-structured plan for training in H-NMR metabolomics and their analysis. Dr. Ambroggio will have on-site training at U Michigan with Dr. Stringer for metabolomics, including sample preparation, NMR spectrum acquisition, spectral processing, metabolite identification and quantification. She will then spend 3 months validating NMR with LC/MS in Dr. Ziady's lab.
- Her research will include specific mentorship for skill sets such as metabolite identification and profiling, latent class modeling, multivariate modeling, and bioinformatics for large scale metabolomics.
- The candidate will participate in a twice-monthly "K club" which is a peer group of faculty and trainees who are applying for or who have received NIH career development awards. This provides camaraderie, mentorship, networking and support for transitioning from a K to R01 funding. She will also participate in a grant writing workshop and a year-long emerging leaders program at U Cincinnati, as well a journal club and a metabolomics seminar.
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Weaknesses

- No weaknesses were noted.

3. Research Plan:

Strengths

- Highly clinically relevant goal of identifying distinct metabolic profiles indicative of viral, typical bacterial, or atypical bacterial CAP infection in children. If successful, this work could substantially focus therapy in children. This would be done in a quantified manner using urine, a non-invasive test, with results and analysis potentially available within hours.
- Important topic of developing a rapid diagnostic test for the etiology of CAP in children. Strong rationale for the need of alternatives to improve diagnostic and therapeutic response. The value of improved diagnostics over poorly performing clinical and radiologic indicators is of heightened importance during a time when antibiotic stewardship is a national priority.
- This work is being performed within the context of a parent study which is now better described. The *Catalyzing Ambulatory Research in Pneumonia Etiology and Diagnostic Innovations in Emergency Medicine* (CARPE DIEM) project will enable Dr. Ambroggio to use extensively collected clinical specimens from study coordinators and clinical data from their electronic medical record. Essentially, the datasets are collected and ready for her in this project. Notably, she herself helped establish the CARPE DIEM project over the past e years, so she is benefitting from her own design. She has strong Emergency Medicine collaborations and a data manager and data programmer for this project.
- She has three important aims
    - 1) to characterize the impact of age and gender on a healthy child's metabolome
    - 2) to compare urine metabolite profiles in non-infected children vs children with confirmed viral vs typical bacterial vs atypical bacterial CAP
    - 3) to determine the impact of antibiotic therapy on serial sampling of urine metabolites in children with and without CAP.

- Data and improved description are now provided for the base study (CARPE DIEM) as requested (distribution of pathogens across 169 children with CAP, allowing for evidence of viral, bacterial, and co-infected (7%) fractions.
- Aim 1 is new and addresses the need to understand normal variation in urine metabolites in healthy children. This was added in direct response to reviewer comments. Preliminary data in 5 healthy children suggests some variation in metabolites in 3 samples, but data from adults suggest that there may be sufficient magnitude of change during infection to overcome these variations. This aim will establish if this is true in children.
- Aim 1 will evaluate urine samples in healthy children at 3 time points D1 (clinic visit), D2 (home), D14 (home) and estimate between subject and within subject variability. Sample size calculation (125 healthy controls) and statistical analysis well-conceived to address variation across host subjects and by gender. Age will also be assessed. This evaluation of age and gender helps balance the choice to use age and gender matched controls in Aim 2 (e.g. aim 2 cannot assess the impact of age or gender)
- Aim 2 preliminary data is much more targeted to the question the candidate is asking. Data now more clear as to the distinction between healthy control children (N=29) and those with viral (N=29) and bacterial pneumonia (N=10) showing at least a 2-fold difference in many metabolites from healthy controls. Larger sample size in the aim has high likelihood for substantiating these data across a range of children and across the CAP types. Definition of CAP by viral and bacterial etiologies remains well conceived and highly clinically relevant, including the use of procalcitonin.
- Sample size (250 cases, 125 controls) is necessary, but nevertheless impressive for this detailed comparison of metabolites as well as the extensive search for a CAP pathogen. Infrastructure in CARPE DIEM is a great strength of this study.
- Aim 3 is an interesting aim of assessing changes in urine metabolites in response to antibiotic therapy to determine if urine can be used as a distinguishing feature to indicate response to antibiotics. Serial collection (D1, D2, D14) wisely mirrors the control collection in Aim 1 for healthy community clinic samples with cases now collected from the ED with CAP. Preliminary data suggest that not only are metabolites different at the diagnosis for CAP vs healthy children, but that some these metabolites change quickly on antibiotics. Interesting exploratory aim.

Weaknesses

- Aim 1 does not specify the study population (age, location). It is assumed it will come from MEEP, but this is not directly stated in the Aim 1 section
- Use of NP swabs as gold standard for CAP etiology does not account for detection of colonizing pathogens (important for the bacterial pathogens or concomitant cold viruses like rhinovirus). Nevertheless, this is the clinical gold standard of diagnostics today.
- It is an important oversight that nowhere in the description of CARPE DIEM does it state whether the diagnosis of CAP requires X ray findings.

4. Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s):

Strengths

- Primary mentor is Dr Samir Shah, a pediatric ID physician who will help provide clinical perspective to her work. Dr. Shah is an expert in pediatric pneumonia and was the lead author of recent national clinical guidelines. He appears to have spent a single year as an Assoc Prof before being offered a Professorship and endowed chair at U Cincinnati.
    - strongly networked in pediatric research multi-institutional consortia.
    - received national research excellence award and a second mentorship award from national pediatric societies in 2015
    - numerous last author papers on pediatric pneumonia; over 200 publications total
    - 40 prior mentees (fellows and junior faculty)
    - funded through April 2017
- Dr. Richard Ruddy is Prof of Pediatrics and nationally known for Emergency Medicine work related to pediatric disease (PECARN). He is the site PI and health system PI for an extensive national pediatric Emergency Medicine research network serially funded through 2019. Well published and funded, has been a co-mentor for Dr. Ambroggio for CARPE DIEM.
- Prof Macaluso is a well-funded long-standing mentor for biostatistics and epidemiologic research and has mentored 15 doctoral students with a high publication record
- Dr. Stringer is a PharmD at U Michigan and director of the metabolomics program. This is one of 6 NIH-funded regional metabolomics facilities. She has mentored over 50 PhD and PharmD candidates. She will provide hands on on-site training for Dr. Ambroggio on metabolomics, including sample preparation, NMR spectrum acquisition, spectral processing, metabolite identification and quantification. She met Dr. Ambroggio at a conference and was impressed by her proactiveness. They co-authored a paper on an approach to a septic child and she provides a strong mentor statement. She is funded through Jan 2020.

Weaknesses

- The primary mentor, Dr. Shah appears to only be funded through Spring 2017 with no pending PI grants. This concern is offset by the fact that the candidate herself is funded to continue the foundational CARPE DIEM project that provides samples for this proposed research.

5. Environment and Institutional Commitment to the Candidate:
Strengths

- Strong institutional commitment. She is already an Assistant Professor and her retention is not dependent on this K award. Divisional funds will help defray the travel costs to U Michigan
- Abundant investment in research by the U of Cincinnati as shown by the internal funds Dr. Ambroggio has already received
- Ample server space provided for datasets and statistical analysis
- Located in close proximity to the offices of mentors

- Fully outfitted NMR and metabolomics laboratories through connection with U Michigan with an NMR spectrometer at U Cincinnati as well
- Largest pediatric research institution in the US in terms of space and third in NIH-funded research. Her primary division (Hospital Medicine) has 45 faculty

Weaknesses

- No weaknesses were noted.

Critique 2

Candidate:	1
Career Development Plan/Career Goals & Objectives:	1
Research Plan:	1
Mentor/Co-Mentor(s), Consultant(s), and Collaborator(s):	1
Environment and Institutional Commitment to the Candidate:	1

Overall Impact:
This is a very strong application that uses metabolomics to investigate and diagnose pediatric CAP. The strengths of the application include the productivity and experience of the principal investigator, the overall design and implementation of the experimental plan, and the quality of the mentors and collaborators that will be involved in the training of Dr. Ambroggio.

1. Candidate:
Strengths

- Excellent record of productivity.
- Highly qualified to conduct the proposed studies.

Weaknesses

- No weaknesses were noted.

2. Career Development Plan/ Career Goals & Objectives:
Strengths

- The proposed coursework is appropriate for the project and will enrich the applicant’s ability to direct future projects.

Weaknesses

- No weaknesses were noted.

3. Research Plan:

Strengths

- The PI has addressed many of the previous concerns dealing with sample size and organization of the specific aims, resulting in an improved research plan.

Weaknesses

- No weaknesses were noted.

4. Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s):

Strengths

- A large and outstanding collection of mentors, consultants and collaborators.

Weaknesses

- No weaknesses were noted.

5. Environment and Institutional Commitment to the Candidate:

Strengths

- The institutional commitment is appropriate and the infrastructure in place is outstanding for this project.

Weaknesses

- No weaknesses were noted.

Critique 3

Candidate:	1
Career Development Plan/Career Goals & Objectives:	1
Research Plan:	2
Mentor/Co-Mentor(s), Consultant(s), and Collaborator(s):	1
Environment and	1

Institutional Commitment to the Candidate:

Overall Impact:
This resubmission is exceptional. The subject area is important and innovative. She fully addresses all of the criticisms identified in the last review. She has published another manuscript since the last submission in the area of CAP raising her grant-relevant track record to 15. Her Career Development Plan is well integrated with her 4 training goals, her mentors' expertise, and her Specific Aims. Her 4 training goals are realistic, pragmatic, and attainable. She has a truly exceptional mentoring team with clearly delineated roles and responsibilities who have demonstrated their commitment to her success by meeting quarterly since 2013. She has largely enrolled her cohort (169 enrolled to date). She provides robust preliminary data supporting the hypotheses and feasibility of each of her 3 Specific Aims.

1. Candidate:
Strengths

- Continued productivity with another 1st author publication since last submission, totaling 19 publications, 10 as 1st author and 15 related to pneumonia
- Involvement as 10 grants, 5 as PI
- 2 new awards for research and mentoring excellence.
- Longstanding dedication to research. Research tech for 3y at Fred Hutchinson led to pursuit of MPH/PhD in Epidemiology at Drexel (2011). Completed post-doc at U Cincinnati and was promoted to Assistant Professor of Pediatrics in 2013, joint appointment in Hospital Medicine and Biostatistics and Epidemiology at Cincinnati Children's Hospital Medical Center.
- Strong letters of recommendation.

Weaknesses

- No weaknesses were noted.

2. Career Development Plan/ Career Goals & Objectives:

Strengths

- Exceptional mentoring plan with specific mentors with distinct roles and specific assignment to completion of specific role
- Includes formal coursework (5) on genomics, medical informatics, advanced stats, and medical microbiology.
- Well-structured plan for training in H-NMR metabolomics and their analysis. Dr. Ambroggio will have on-site training at U Michigan with Dr. Stringer for metabolomics, including sample preparation, NMR spectrum acquisition, spectral processing, metabolite identification and quantification. She will then spend 3 months validating NMR with LC/MS in Dr. Ziady's lab. 4 Training goals that relate directly to her current science: 1) quantitative NMR metabolite generation and analysis, 2) advanced statistical analyses; 3) metabolomic data

interpretation; and 4) professional skills development. Table 2 beautifully maps out how her planned academic courses and experiential learning contribute to these goals.

- Longstanding mentoring committee with quarterly meetings since 2013. Will continue weekly meetings with Dr. Shah as her primary mentor, and every other week calls with Dr. Stringer, her mentor for metabolomics. She will have monthly meetings with her co-mentor on molecular epi, and monthly meetings with Shah/Ruddy/Florin in development of a pediatric cohort on which her prelim data are based.
- Her research will include specific mentorship for skill sets such as metabolite identification and profiling, latent class modeling, multivariate modeling, and bioinformatics for large scale metabolomics.
- The candidate will participate in a twice-monthly “K club,” which is a peer group of faculty and trainees who are applying for or who have received NIH career development awards.

Weaknesses

- No weaknesses were noted.

3. Research Plan:
Strengths

- PI has fully resolved all prior criticisms of the Research Plan. She has added a specific aim to confirm baseline metabolomic data for healthy controls and provides additional preliminary data to support all 3 of her SA since last review.
- Highly clinically relevant goal of identifying distinct metabolic profiles indicative of viral, typical bacterial, or atypical bacterial CAP infection in children. If successful, this work could substantially focus therapy in children. This would be done in a quantified manner using urine, a non-invasive test, with results and analysis potentially available within hours and offers the long-term prospect of being able to avoid antibiotic administration to the 60% of CARPE-DIEM patients with viral infection.
- Important topic of developing a rapid diagnostic test for the etiology of CAP in children. Strong rationale for the need of alternatives to improve diagnostic and therapeutic response. The value of improved diagnostics over poorly performing clinical and radiologic indicators is of heightened importance during a time when antibiotic stewardship is a national priority.
- PI now provides more specifics as to her cohort. For SA1, she now provides a demographics table for the 169 patients from CARPE-DIEM with urine samples, reassuring as to the availability of existing samples. In addition, she provides convenience sample data from 5 patients demonstrating changes in metabolite concentrations over time, confirming the platform capacity to conduct the proposed experiments proposed in SA1.
- For SA2 preliminary data, PI now provides a table outlining the etiology of CAP in her MEEP cohort of 169 patients. It is interesting that an adequate number of subjects have exclusively viral (n=102) pneumonia. Bacterial monomicrobial etiology is somewhat more modest (n=21).
- For SA2, preliminary data is also added that demonstrates PLS-DA score plot differentiation of bacterial vs healthy and viral vs. healthy subjects (Figure 4). In addition, she now

provides a radar plot of quantitative NMR metabolomics of urine from pediatric patients with pneumonia due to bacteria or virus and healthy controls.

- SA3 preliminary data now provides changes in metabolite concentrations over time that demonstrates clear differences in controls and pneumonia subjects..
- CARPE DIEM the source of her clinical ascertainment. It is externally funded and fully operational
- Case definitions are reasonable.
- Budget justification provides further strong evidence as to the attention that PI has invested upon the administration of the project. For example, she includes significant personnel support for data management of the dataset.
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Weaknesses

- Bacterial monomicrobial etiology cohort is small. This could be addressed by enriching the cohort for cases of bacterial CAP by high-risk enrolling or selective ascertainment from hospitalized subjects, if PI feels this is a potential risk.
- The fact that only 20% suggests that some of the cases attributed to viral or bacterial etiology might be false positives based upon the recent NEJM multicohort center enrollment of etiologies of adults with CAP.
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4. Mentor(s), Co-Mentor(s), Consultant(s), Collaborator(s):
Strengths

- PI has fully addressed all concerns related to mentors and collaborators. The large panel of mentors now have clearly delineated roles that PI now outlines in Tables 1 & 2.
- NMR PI has new R01 in related topic of NMR analysis in sepsis that could potentially provide beneficial information for Ambroggio's K01.
- Primary mentor is Dr. Samir Shah, a pediatric ID physician who will help provide clinical perspective to her work. Dr. Shah is an expert in pediatric pneumonia and was the lead author of recent national clinical guidelines. He appears to have spent a single year as an Assoc Prof before being offered a Professorship and endowed chair at U Cincinnati.strongly networked in pediatric research multi-institutional consortia.
    - *numerous last author papers on pediatric pneumonia.*
    - *40 prior mentees (fellows and junior faculty).*
    - *funded through 2017.*
    -
- Prof Macaluso is a well-funded long-standing mentor for biostatistics and epidemiologic research and has mentored 15 doctoral students with a high publication record.
- Dr. Stringer is a PharmD at U Michigan and director of the metabolomics program. She has mentored over 50 PhD and PharmD candidates. She will provide hands on training for Dr. Ambroggio on metabolomics, including sample preparation, NMR spectrum acquisition, spectral processing, metabolite identification and quantification. She met Dr. Ambroggio at a conference and was impressed by how proactive she was. They co-authored a paper on an approach to a septic child and she provides a strong mentor statement.

Weaknesses

- No weaknesses were noted.

5. Environment and Institutional Commitment to the Candidate:
Strengths

- Strong institutional commitment. She is already an Assistant Professor and her retention is not dependent on this K award. Divisional funds will help defray the travel costs to U Michigan.
- Abundant investment in research by the U of Cincinnati as shown by the internal funds Dr. Ambroggio has already received.
- Ample server space provided for datasets and statistical analysis.
- Located in close proximity to the offices of mentors.
- Fully outfitted NMR and metabolomics laboratories through connection with U Michigan with an NMR spectrometer at U Cincinnati as well.

Weaknesses

- No weaknesses were noted.

THE FOLLOWING RESUME SECTIONS WERE PREPARED BY THE SCIENTIFIC REVIEW OFFICER TO SUMMARIZE THE OUTCOME OF DISCUSSIONS OF THE REVIEW COMMITTEE ON THE FOLLOWING ISSUES:

PROTECTION OF HUMAN SUBJECTS (Resume): Code: 30 ACCEPTABLE

The application adequately addresses these four points: Risks to human subjects; Adequacy of protections against these risks; Potential benefits of the proposed research to human subjects; and Importance of the knowledge to be gained from this project.

Projects to be undertaken with support of this training grant, which involve human subjects, must conform to the NIH policies on the protection of human subjects. Guidance can be found in PHS398 application materials and the NIH Office of Extramural Research web site http://grants.nih.gov/grants/policy/hs/index.htm.

DATA AND SAFETY MONITORING PLAN: ACCEPTABLE

Comments or Concerns: Concur no need for DSMB. The PI will monitor for anticipated and unanticipated issues.

INCLUSION OF WOMEN PLAN (Resume): Code: G1A ACCEPTABLE
Based upon the known demographics in the Emergency Department at CCHMC and the Fairfield Primary Care Clinic, the study population for both aims will be approximately 48% female.

Projects to be undertaken with support from this training grant, which involve clinical research studies, must conform to the NIH policies on the inclusion of women in study populations. See http://grants.nih.gov/grants/funding/women_min/guidelines_amended_10_2001.htm. Details of such studies, including a description of the population and rationale for inclusion/exclusion, must be provided to the NIH program administrator of this training grant prior to undertaking the studies.

INCLUSION OF MINORITIES PLAN (Resume): Code: M1A ACCEPTABLE
Based upon the known demographics in the Emergency Department at CCHMC and the Fairfield Primary Care Clinic, the study population for both aims will be approximately 48% white, 45% black or African American and 96% identifying as non-Hispanic ethnicity.

Projects to be undertaken with support from this training grant, which involve clinical research studies, must conform to the NIH policies on the inclusion of minorities in study populations.
See http://grants.nih.gov/grants/funding/women_min/guidelines_amended_10_2001.htm. Details of such studies, including a description of the population and rationale for inclusion/exclusion, must be provided to the NIH program administrator of this training grant prior to undertaking the studies.

INCLUSION OF CHILDREN PLAN (Resume): Code: C2A ACCEPTABLE
Only children, 3 months to 11 years of age will be included in this investigation.

Projects to be undertaken with support from this training grant, which involve clinical research studies, must conform to the NIH policies on the inclusion of children in study populations. See http://grants.nih.gov/grants/funding/children/children.htm. Details of such studies, including a description of the population and rationale for inclusion/exclusion, must be provided to the NIH program administrator of this training grant prior to undertaking the studies.

VERTEBRATE ANIMALS (Resume): Code: 10 NOT APPLICABLE
No vertebrate animals will be used in the performance of this work.

BIOHAZARD COMMENT: NOT APPLICABLE
No biohazards were identified.

RESUBMISSION:
Comments: See above reviewer comments. All issues were addressed.

TRAINING IN THE RESPONSIBLE CONDUCT OF RESEARCH: ACCEPTABLE

Comments:

Format: formal ethics in research course plus seminars; previously completed a 2 10-week course training in research ethics and CITI training. Will also be completing all IRB protocols.

Subject Matter: IRB regulations, human subjects, consent, special populations

Faculty Participation: active participants in responsible conduct of research conference

Duration: Seminar (24 hours over 5y), symposium (8 hours every two years), conference once per month with faculty on responsible conduct of research; coursework (10 hours over 5y)

Frequency: monthly faculty conference, and annual symposium, periodic selection of seminars

FOREIGN INSTITUTION: NOT APPLICABLE

SELECT AGENTS: NOT APPLICABLE

RESOURCE SHARING PLANS: NOT APPLICABLE

Data Sharing Plan Comments (if >$500,000/year):

Sharing Model Organisms Comments:

Genomic Data Sharing (GDS) Comments:

AUTHENTICATION OF KEY BIOLOGICAL AND/OR CHEMICAL RESOURCES: UNACCEPTABLE
A separate 1 page “Authentication” document using the new NIH Forms D should be uploaded with the application. See Implementing Rigor and Transparency in NIH & AHRQ Career Development Award Applications http://grants.nih.gov/grants/guide/notice-files/NOT-OD-16-012.html.

BUDGET AND PERIOD OF SUPPORT: The budget was recommended as requested.

Footnotes for 1 K01 AI125413-01A1; PI Name: Ambroggio, Lilliam

NIH has modified its policy regarding the receipt of resubmissions (amended applications). See Guide Notice NOT-OD-14-074 at http://grants.nih.gov/grants/guide/notice-files/NOT-OD-14-074.html. The impact/priority score is calculated after discussion of an application by averaging the overall scores (1-9) given by all voting reviewers on the committee and multiplying by 10. The criterion scores are submitted prior to the meeting by the individual reviewers assigned to an application, and are not discussed specifically at the review meeting or calculated into the overall impact score. Some applications also receive a percentile ranking. For details on the review process, see http://grants.nih.gov/grants/peer_review_process.htm#scoring.