SUMMARY STATEMENT

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(Privileged Communication)

Release Date: **07/10/2018**
Revised Date:

Application Number: **1 F30 MH118865-01**

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Review Group:	**ZRG1 F02B-D (20)** **Center for Scientific Review Special Emphasis Panel** **Fellowships: Sensory and Motor Neuroscience, Cognition and Perception**
Meeting Date:	**06/21/2018**
Council:	**OCT 2018** ***PCC:*** **7K-TGMC2**
Requested Start:	**09/01/2018**

Project Title:	**Sources of Cholinergic Modulation of Cortical Microcircuits**
	***Requested*: 4 Years**
Sponsor:	**BARTH, ALISON L**
Department:	**Biological Sciences**
Organization:	**CARNEGIE-MELLON UNIVERSITY**
City, State:	**PITTSBURGH PENNSYLVANIA**
SRG Action:	**Impact Score:30 Percentile:23**
Next Steps:	**Visit https://grants.nih.gov/grants/next_steps.htm**
Human Subjects:	**10-No human subjects involved**
Animal Subjects:	**30-Vertebrate animals involved - no SRG concerns noted**

1F30MH118865-01 Myal, Stephanie

RESUME AND SUMMARY OF DISCUSSION:
This predoctoral MD/PhD fellowship application proposes training while characterizing the cholinergic modulation of mouse somatosensory cortical circuits using anatomical, optogenetic and electrophysiological approaches. The candidate was viewed as exceptionally strong with significant previous research experience in multiple labs, coauthorships on 7 publications, highly supportive letters, and appropriate personal training goals. The sponsor has excellent experience, technical expertise, and high productivity. She has a successful record as a mentor and available funding, and her training plan for the candidate is thoughtful, detailed and individualized. Notably, the candidate already has a co-authorship on a Neuron paper with the sponsor. Carnegie Mellon provides ample facilities and resources for medical scientist training. Reviewers were generally positive about the questions being addressed in the research plan which presents clear hypotheses that are predicated on the sponsor's previous work, and the investigation of a fast-acting, location-specific role for acetylcholine in the cortex was viewed as exciting. The two sets of experiments proposed are cohesive, complementary, and feasible, and potential pitfalls and alternative outcomes are considered. The panel discussed a few weaknesses, however: some felt that the rationale for the focus on somatosensory cortex was not particularly well explained, that there was not a satisfying conceptual justification for generalizing results in mice to broader principles about brain function e.g. in primates, and controls for optogenetic stimulation were not fully considered. Although the plan is ambitious, the candidate appears to have already mastered many of the techniques involved, and the panel saw very strong training potential for the candidate to continue to advance her goal of conducting translational research in neural circuits that underlie cognitive function and dysfunction. After discussion, with reviewers weighing the merits of the candidate, sponsor, and training potential against minor to moderate weaknesses in the research plan, final ratings were mostly high with a few in the moderate range for this predoctoral training application and the foundation it would provide for a future career as a clinician scientist.

DESCRIPTION (provided by applicant): The cognitive symptoms of schizophrenia are subtle, persistent, debilitating, and more strongly linked to treatment non-adherence and poor prognosis than the overt, better-known psychotic symptoms. They are also poorly treated by antipsychotics. A large body of indirect evidence implicates deficient signaling of acetylcholine (ACh) in schizophrenia cognitive symptoms. ACh normally acts as an attention signal, activating a chain of GABA-ergic inhibition in the cerebral cortex that increases the activity of output neurons. In our published work, we see that ACh acts in a precise manner to strengthen specific synapses in the mouse somatosensory cortex (S1). Preliminarily, we see that different cell types respond distinctly to ACh, mediated by nicotinic and muscarinic ACh receptors. We are interested in how ACh from different sources affects cortical circuits, and whether ACh release sites are preferentially associated with specific intra-cortical cell types and thalamocortical synapses. The main source of cortical ACh is the basal forebrain, but there are also intrinsic cortical ACh neurons of unknown function which may augment ACh signaling in a precise manner. In Aim 1.1, we propose to anatomically test the relative contribution of extrinsic and intrinsic sources of ACh inputs using immunolabeling of ACh release sites for advanced confocal imaging and 3D reconstructions of ACh release sites. In Aim 1.2, we will functionally assess the contribution of these sources in whole-cell recordings, using optogenetics to evoke ACh release in brain slices. We hypothesize that endogenous ACh has cell type- and layer-specific responses differentially controlled by intrinsic and extrinsic ACh sources. We also suspect that the precise localization of ACh release sites contributes to synapse-specific thalamocortical effects of ACh. In our recent paper, we found that endogenous ACh strengthened excitatory synapses onto particular interneuron types via presynaptic mechanisms. However, when we activated cholinergic receptors pharmacologically, both synapses were strengthened, suggesting that ACh receptors and/or release sites are spatially segregated to precisely tune excitation. Prior pharmacological work suggests that thalamic inputs from the ventral

posterior medial nucleus (VPM, a lower-order sensory thalamic nucleus) express nicotinic receptors, but it is unknown whether these inputs would be strengthened by endogenous ACh, or whether inputs from a higher order sensory thalamic nucleus (the posterior medial nucleus, POm) are also influenced by ACh. In Aim 2.1, we will anatomically compare whether ACh release sites are more common at VPM or POm synapses. In Aim 2.2 we will functionally assess whether endogenous ACh strengthens POm and VPM inputs using whole-cell recordings with dual-color optogenetics in brain slices. We hypothesize that ACh release sites are associated with POm inputs, and that timed endogenous ACh enhances thalamocortical inputs onto specific cell types, potentially shifting cortical circuits to favor different thalamic information streams during states of attention or disease-related cholinergic dysfunction.

PUBLIC HEALTH RELEVANCE: Schizophrenia is characterized by overt psychosis, but the subtler cognitive symptoms have a greater impact on quality of life and prognosis and are poorly treated by antipsychotics. Acetylcholine, a neurotransmitter important for cognition and attention, fine-tunes cortical circuits to achieve a complex balance of excitation and inhibition that is disrupted in schizophrenia. The proposed work will test the effects of acetylcholine from diverse sources on neural circuits in the mouse somatosensory cortex, and will inform the ongoing development of cholinergic therapies for cognitive impairment in schizophrenia and other psychiatric diseases.

CRITIQUE 1

Fellowship Applicant: 1
Sponsors, Collaborators, and Consultants: 2
Research Training Plan: 2
Training Potential: 1
Institutional Environment & Commitment to Training: 3

Overall Impact/Merit*:* This training proposal is concerned with characterizing the source of cortical cholinergic innervation and determining if acetylcholine enhances thalamocortical transmission. The overall score is based on the exceptional qualifications of the applicant and the quality of the training plan and its potential for providing a good foundation for a career as a clinical research scientist. The applicant has extensive research experience and has achieved a long list of accomplishments that include authorship on 7 research papers. The training plan is well designed. The research plan consists of experiments that systematically analyze the anatomical basis of cholinergic innervation in the cortex and is followed by studies examining the physiological effects of acetylcholine on thalamocortical transmission. This is a significant scientific and clinical problem that has implications for the treatment of Alzheimer's disease and other neurocognitive diseases, and the overall training program overlaps with the applicant's goal of conducting translational research that will impact our understanding of the neural basis of cognition and its demise in certain psychiatric conditions. No significant weaknesses could be identified in this proposal.

1. Fellowship Applicant:

Strengths

- The applicant has a very strong undergraduate academic record with virtually all A's in a premedical curriculum.
- Applicant acquired extensive neuroscience research experience examining neural circuit mechanisms and their influence on behavior at University of Maryland, University of Arkansas, and the National Institute of Drug Abuse.
- The applicant has established an impressive list of awards and honors

- The applicant's strong commitment to neuroscience research and medicine is evident from her participation in numerous activities including her involvement in assessing the neuropsychological outcomes of DBS, her participation with a neurosurgeon in developing a business model for a chronic pain tracker, and her activities in medical education.
- The applicant has already acquired research experience that is directly relevant to the present proposal including whole cell recordings in brain slices, optogenetics, and cholinergic neuropharmacology.
- The applicant has a strong background in the research enterprise including the practice of scientific communication as indicated by her authorship on 7 peer-reviewed publications and numerous abstracts and poster presentations at several scientific conferences.
- The letters of recommendation are extremely strong; they all agree that the applicant is intelligent, strategic in her thinking, and have uniformly expressed astonishment at her unusual high level of achievement and productivity.

Weaknesses

- None noted.

2. Sponsors, Collaborators, and Consultants:

Strengths

- Dr. Alison Barth, the sponsor, is an outstanding neuroscientist with extensive experience using whole-cell patch clamp electrophysiology, neuronal tracing, cellular identification, and other techniques relevant to this proposal.
- Dr. Barth has a productive research record; she has published more than 10 papers in the last three years alone, including several using the techniques described in this proposal.
- Dr. Barth has previously trained a half dozen predoctoral students and 4 postdoctoral associates in her lab, most of which have successfully progressed in their scientific careers.
- The applicant did research rotations as a MSTP student, and the selection of Dr. Barth as lab sponsor appears to represent a good match with the applicant's research interests.
- Dr. Barth is PI on three active research grants, including two that will extend into 2020.
- Dr. Barth provided a comprehensive description of the training plan and the applicant has already established a publication track record with the sponsor with a co-authored paper in Neuron.

Weaknesses

- None noted.

3. Research Training Plan:

Strengths

- The research plan is based on two cohesive set of experiments. Experiments concerned with characterizing the source of cholinergic innervation of cortical neurons (basal forebrain vs cortical VIP neurons) are nicely complemented by experiments designed to determine if these sources enhance thalamocortical transmission.
- The series of proposed experiments represent a well-designed systematic approach in examining the cholinergic innervation and physiological effects of endogenous acetylcholine on cortical pyramidal neurons and three different subtypes of interneurons.

- The research plan is feasible and consideration was given to alternative outcomes and pitfalls
- Cholinergic innervation of cortical circuitry is a significant scientific and clinical problem that matches well with the applicant's goal of conducting translational research that will advance our understanding of the neural circuits responsible for mediating cognition and its demise in certain psychiatric syndromes and neurodegenerative diseases.

Weaknesses

- As described in the research strategy and as illustrated in Figure 4, the quantification of cholinergic terminals on specific cells appears to be possible with high-resolution confocal microscopy, but confidence in the resulting data might be strengthened by validation with ultrastructural evidence.

4. Training Potential:

Strengths

- The applicant interacts with several advanced trainees in the Barth lab who will provide technical and intellectual support.
- The applicant has acquired skills in brain slice electrophysiology, and will expand her technical abilities in dual-color optogenetic stimulation, confocal microscopy, cholinergic neuropharmacology, and data analysis. In developing these skills, the applicant has access to a number of experts with knowledge and experience in these areas.
- The proposed research seems likely to lead to a number of high quality publications
- The sponsor has clearly described a comprehensive training plan that includes numerous professional development opportunities on a regular basis (weekly and monthly) that should enhance the applicant's training in data analysis, scientific communication, grant writing, and continue to develop the applicant's knowledge of contemporary issues in neuroscience.
- The training plan also includes individual supervision and regular interactions between the sponsor and the applicant, as well as formal interactions with the applicant's thesis committee and the MSTP advisory committee.
- Overall, the training plan appears to be ideal for providing a solid foundation for an independent research career as a clinical research scientist

Weaknesses

- None noted.

5. Institutional Environment & Commitment to Training:

Strengths

- The Institution provides a number of seminars, regular workshops, and other opportunities to enhance the applicant's training.
- The applicant has already taken institutional MSTP courses and related workshops focused on Professional Development, Scientific Writing, Reproducibility in Science, Biomedical Ethics, as well as activities and instruction aimed at integrating clinical responsibilities with scientific research
- The facilities and resources needed for the proposed research are provided by the sponsor's laboratory and the research cores (e.g., breeding facility) of the institution.

Weaknesses

- None noted.

Protections for Human Subjects:

Not Applicable (No Human Subjects)

Vertebrate Animals:

YES, all criteria addressed

- the use and treatment of animals is appropriate

Biohazards:

Not Applicable (No Biohazards)

Training in the Responsible Conduct of Research:

Acceptable

Comments on Format (Required):

- multiple academic units provided the applicant with RCR training

Comments on Subject Matter (Required):

- appropriate

Comments on Faculty Participation (Required):

- RCR training is also discussed and provided by mentor

Comments on Duration (Required):

- 10 week MSTP course, 8 week course in CMU Biology program

Comments on Frequency (Required):

- repeated ongoing discussions with mentor in lab meetings and journal clubs

Resource Sharing Plans:

Acceptable

- All experiments and results are freely shared

Budget and Period of Support:

Recommend as Requested

CRITIQUE 2

Fellowship Applicant: 1
Sponsors, Collaborators, and Consultants: 2
Research Training Plan: 2
Training Potential: 2
Institutional Environment & Commitment to Training: 2

Overall Impact/Merit: This is an application for support during MD/PhD training by a very impressive applicant. She is currently a 2nd year PhD student and 4th year MD (MSTP). Four years additional support are requested. The applicant graduated (at age 19) from Arkansas State University with a double major in biology and chemistry and excellent grades. She continues to obtain excellent grades in graduate school. She already sought out research opportunities as an undergraduate and then worked for four years as a research tech which resulted in co-authorships on several publications. The sponsor is an expert in the field of study proposed here and he has a strong training record and has secured good funding. The chosen research project is of high significance. It is ambitious but even partial completion will lead to good publications with a high impact. The training potential for this applicant comprises learning additional new techniques, of which she already masters many, and also taking charge of a project and transitioning towards increased independence for the next stages of her career. Overall, this is an excellent proposal.

1. Fellowship Applicant:

Strengths

- Straight A undergraduate, similar graduate school grades
- Three years research experience as undergraduate, included mentoring other undergraduate and MS students
- Four years as research tech a U. Arkansas, SoM NIDA, U Maryland SoM (a section “Position and Honors” in the biosketch would have been helpful), resulting in several co-authorships
- Overall, co-author of 7 journal publications, including one (Neuron) with sponsor which is basis for thesis topic. Also first or co-author on several abstracts
- All required coursework and teaching requirements done, passed qualifying exam

Weaknesses

- Articles in preparation should not be listed in the biosketch

2. Sponsors, Collaborators, and Consultants:

Strengths

- Dr. Barth is a Professor of Biological Sciences at CMU and an expert in the thesis topic
- Well funded, PI on one R01 and R21, CoPI on another R01 (expiring between 9/19 and 8/20)
- Has trained 6 PhD students and 5 postdocs. All listed have continued good careers, mostly in academia. It is unclear why outcome for one of the postdocs is not listed.

Weaknesses

- None noted

3. Research Training Plan:

Strengths

- Going beyond the “bath application” view of the function of neuromodulators, in particular ACh, by taking into account neuronal and synaptic specificity is of high importance for understanding neural function and also likely clinically. The proposal questions, with good reasons, the idea that “partner less” ACh receptor s are randomly distributed. They will test the hypothesis that, instead, they are localized in the vicinity of particular layers, cell types and subcellular

compartments, and therefore cause differentiated responses (Aim 1). Furthermore, these may differ between cholinergic input from basal forebrain and cortical sources.

- There is very little work on cortical sources of ACh which include the putative cholinergic VIP neurons. Their role needs to be understood
- A similar analysis is applied to thalamocortical projections (Aim 2) which have been very little studied. The sponsor has a recent paper (ref 114) showing her lab can target the relevant thalamic nuclei and their specific cortical projections
- Likewise important is the understanding of this function at higher time resolution (down to ms) than traditionally studied (seconds to minutes).
- The double color optogenetic method to be used will be a new technique for the applicant
- Potential problems are addressed

Weaknesses

- This is an ambitious research program given that the applicant has to fulfill other MSTP requirements during the fellowship (clerkship etc). However, there is not much interdependence between the different components and even partial completion will lead to publications with high impact

4. Training Potential:

Strengths

- The applicant is already trained in most (not all) of the techniques she will apply but she will now learn to be truly in charge of the project, rather than in a supporting role. Her expressed goal of published journal papers as first author (which she has not done yet) shows that she is aware of the needed change in her role

Weaknesses

- None noted.

5. Institutional Environment & Commitment to Training:

Strengths

- UPMC and the Pitt-CMU Center for the Neural Basis of Cognition provide a great environment for neuroscience research
- All equipment needed is in place
- There is one additional PhD students and 3 postdocs (a fourth to join in the spring) in the lab. The other student will graduate soon and the sponsor intends to recruit two additional predoctoral students. This will allow for interactions between students and also sufficient personal attention by the sponsor

Weaknesses

- None noted.

Protections for Human Subjects:

Not Applicable (No Human Subjects)

Vertebrate Animals:

YES, all criteria addressed

- □□□Sponsor is highly experienced in this kind of work□□

Biohazards:

Not Applicable (No Biohazards)

Training in the Responsible Conduct of Research:

Acceptable

Comments on Format (Required):

- No course on RCR is listed. However, the sum of all the activities listed that provide training in RCR from different angles is likely acceptable□□□□

Comments on Subject Matter (Required):

- □Depending on classes□□□□

Comments on Faculty Participation (Required):

- □All listed have faculty participation□□□□

Comments on Duration (Required):

- □Varies by classes□□

Comments on Frequency (Required):

- □□□Varies by classes□□

Applications from Foreign Organizations:

Not Applicable

Resource Sharing Plans:

Acceptable

- □□□Dissemination through articles. If software is generated, it will be shared.

Budget and Period of Support:

Recommend as Requested

Recommended budget modifications or possible overlap identified:

- □□□Potential overlap noted with NIH RF1MH114103□

CRITIQUE 3

Fellowship Applicant: 1
Sponsors, Collaborators, and Consultants: 2
Research Training Plan: 5
Training Potential: 3
Institutional Environment & Commitment to Training: 2

Overall Impact/Merit: This is a good application by an excellent applicant who will undertake studies that examine how ACh from different sources affects cortical circuits, and if release sites are associated with specific cell types in the cortex and the thalamus. Both the sponsor and the environment are excellent and the training potential is very high. Unfortunately, the application lacked coherence, was extremely difficult to read, and the significance of these studies using this model was not well justified.

1. Fellowship Applicant:

Strengths

- Excellent letters
- Outstanding academic record and publications

Weaknesses

- None noted.

2. Sponsors, Collaborators, and Consultants:

Strengths

- Well-funded
- Good record of publications
- Excellent consultants
- Very good record of training graduate students and post docs

Weaknesses

- None noted.

3. Research Training Plan:

Strengths

- None noted.

Weaknesses

- Extremely dense and difficult to read
- It looks like they will be examining S1, but there is no justification for this choice or details on how they will validate where they are in the neocortex
- Never really justified how these experiments in mice could be extrapolated to humans.

4. Training Potential:

Strengths

- Detailed training plan
- Will learn a variety of new techniques including patch clamp electrophysiology, optogenetics and anatomically targeted viral injections

Weaknesses

- None noted.

5. Institutional Environment & Commitment to Training:

Strengths

- excellent

Weaknesses

- None noted.

Protections for Human Subjects:

Not Applicable (No Human Subjects)

Vertebrate Animals:

YES, all criteria addressed

Biohazards:

Not Applicable (No Biohazards)

Training in the Responsible Conduct of Research:

Acceptable

Comments on Format (Required): acceptable

Comments on Subject Matter (Required): acceptable

Comments on Faculty Participation (Required): acceptable

Comments on Duration (Required): acceptable

Comments on Frequency (Required): acceptable

Select Agents:

Acceptable

Resource Sharing Plans:

Acceptable

Budget and Period of Support:

Recommend as Requested

THE FOLLOWING SECTIONS WERE PREPARED BY THE SCIENTIFIC REVIEW OFFICER TO SUMMARIZE THE OUTCOME OF DISCUSSIONS OF THE REVIEW COMMITTEE, OR REVIEWERS' WRITTEN CRITIQUES, ON THE FOLLOWING ISSUES:

VERTEBRATE ANIMALS: ACCEPTABLE

COMMITTEE BUDGET RECOMMENDATIONS: The budget was recommended as requested.

Footnotes for 1 F30 MH118865-01; PI Name: Myal, Stephanie E.

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.

MEETING ROSTER

Center for Scientific Review Special Emphasis Panel
CENTER FOR SCIENTIFIC REVIEW
Fellowships: Sensory and Motor Neuroscience, Cognition and Perception
ZRG1 F02B-D (20)
06/21/2018 - 06/22/2018

Notice of NIH Policy to All Applicants: Meeting rosters are provided for information purposes only. Applicant investigators and institutional officials must not communicate directly with study section members about an application before or after the review. Failure to observe this policy will create a serious breach of integrity in the peer review process, and may lead to actions outlined in NOT-OD-14-073 at https://grants.nih.gov/grants/guide/notice-files/NOT-OD-14-073.html and NOT-OD-15-106 at https://grants.nih.gov/grants/guide/notice-files/NOT-OD-15-106.html, including removal of the application from immediate review.

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