

PI: FROEMKE, ROBERT CROOKS	Title: Synaptic basis of perceptual learning in primary auditory cortex	
Received: 07/05/2016	Opportunity: PA-16-160	Council: 01/2017
Competition ID: FORMS-D	FOA Title: NIH Research Project Grant (Parent R01)	
2R01DC012557-06	Dual: MH,LM	Accession Number: 3954637
IPF: 5998304	Organization: [REDACTED]	
Former Number:	Department: Otolaryngology	
IRG/SRG: ZRG1 AUD-Z (90)S	AIDS: N	Expedited: N
<u>Subtotal Direct Costs</u> (excludes consortium F&A) Year 6: [REDACTED] [REDACTED] [REDACTED] [REDACTED] [REDACTED] [REDACTED] [REDACTED] [REDACTED] [REDACTED]	Animals: Y Humans: N Clinical Trial: N Current HS Code: 10 HESC: N	New Investigator: N Early Stage Investigator: N
Senior/Key Personnel:	Organization:	Role Category:
Robert Froemke	[REDACTED]	PD/PI

Additions for Review

Accepted Publication

Froemke.pdf

APPLICATION FOR FEDERAL ASSISTANCE
SF 424 (R&R)

		3. DATE RECEIVED BY STATE	State Application Identifier
1. TYPE OF SUBMISSION*		4.a. Federal Identifier DC012557	
<input type="radio"/> Pre-application <input checked="" type="radio"/> Application <input type="radio"/> Changed/Corrected Application		b. Agency Routing Number	
2. DATE SUBMITTED	Application Identifier	c. Previous Grants.gov Tracking Number	
5. APPLICANT INFORMATION		Organizational DUNS*: [REDACTED]	
Legal Name*:	[REDACTED]		
Department:	Sponsored Programs Admin		
Division:			
Street1*:	[REDACTED]		
[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]		
Province:			
Country*:	USA: UNITED STATES		
ZIP / Postal Code*:	[REDACTED]		
Person to be contacted on matters involving this application			
Prefix:	First Name*: Irfana	Middle Name:	Last Name*: Ahmed
			Suffix:
Position/Title:	Grants Specialist		
Street1*:	[REDACTED]		
[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]		
County:			
State*:	[REDACTED]		
Province:			
Country*:	USA: UNITED STATES		
ZIP / Postal Code*:	[REDACTED]		
Phone Number*:	[REDACTED]	Fax Number:	[REDACTED]
		Email:	[REDACTED]
6. EMPLOYER IDENTIFICATION NUMBER (EIN) or (TIN)* [REDACTED]			
7. TYPE OF APPLICANT*		O: Private Institution of Higher Education	
Other (Specify):			
Small Business Organization Type		<input type="radio"/> Women Owned <input type="radio"/> Socially and Economically Disadvantaged	
8. TYPE OF APPLICATION*		If Revision, mark appropriate box(es).	
<input type="radio"/> New <input type="radio"/> Resubmission <input checked="" type="radio"/> Renewal <input type="radio"/> Continuation <input type="radio"/> Revision		<input type="radio"/> A. Increase Award <input type="radio"/> B. Decrease Award <input type="radio"/> C. Increase Duration <input type="radio"/> D. Decrease Duration <input type="radio"/> E. Other (specify) :	
Is this application being submitted to other agencies?* <input type="radio"/> Yes <input checked="" type="radio"/> No What other Agencies?			
9. NAME OF FEDERAL AGENCY* National Institutes of Health		10. CATALOG OF FEDERAL DOMESTIC ASSISTANCE NUMBER TITLE:	
11. DESCRIPTIVE TITLE OF APPLICANT'S PROJECT* Synaptic basis of perceptual learning in primary auditory cortex			
12. PROPOSED PROJECT		13. CONGRESSIONAL DISTRICTS OF APPLICANT	
Start Date*	Ending Date*	[REDACTED]	
04/01/2017	03/31/2022		

14. PROJECT DIRECTOR/PRINCIPAL INVESTIGATOR CONTACT INFORMATION

Prefix: First Name*: Robert Middle Name: C Last Name*: Froemke Suffix:
 Position/Title: Assistant Professor
 Organization Name*: [REDACTED]
 Department: Otolaryngology
 Division: [REDACTED]
 Street2: [REDACTED]
 City*: [REDACTED]
 County: [REDACTED]
 State*: [REDACTED]
 Province: [REDACTED]
 Country*: USA: UNITED STATES
 ZIP / Postal Code*: [REDACTED]
 Phone Number*: [REDACTED] Fax Number: [REDACTED] Email*: [REDACTED]

15. ESTIMATED PROJECT FUNDING

a. Total Federal Funds Requested*

[REDACTED]
 [REDACTED]
 [REDACTED]

16. IS APPLICATION SUBJECT TO REVIEW BY STATE EXECUTIVE ORDER 12372 PROCESS?*

a. YES THIS PREAPPLICATION/APPLICATION WAS MADE AVAILABLE TO THE STATE EXECUTIVE ORDER 12372 PROCESS FOR REVIEW ON:

DATE:

b. NO PROGRAM IS NOT COVERED BY E.O. 12372; OR
 PROGRAM HAS NOT BEEN SELECTED BY STATE FOR REVIEW

17. By signing this application, I certify (1) to the statements contained in the list of certifications* and (2) that the statements herein are true, complete and accurate to the best of my knowledge. I also provide the required assurances * and agree to comply with any resulting terms if I accept an award. I am aware that any false, fictitious, or fraudulent statements or claims may subject me to criminal, civil, or administrative penalties. (U.S. Code, Title 18, Section 1001)

I agree*

* The list of certifications and assurances, or an Internet site where you may obtain this list, is contained in the announcement or agency specific instructions.

18. SFLLL or OTHER EXPLANATORY DOCUMENTATION

File Name:

19. AUTHORIZED REPRESENTATIVE

Prefix: First Name*: Anthony Middle Name: R Last Name*: Carna Suffix:
 Position/Title*: Sr Dir-Sponsored Prog Adm(SPA)
 Organization Name*: [REDACTED]
 Department: Sponsored Programs Admin
 Division: [REDACTED]
 Street1*: [REDACTED]
 County: [REDACTED]
 State*: [REDACTED]
 Province: [REDACTED]
 Country*: USA: UNITED STATES
 ZIP / Postal Code*: [REDACTED]
 Phone Number*: [REDACTED] Fax Number: [REDACTED] Email*: [REDACTED]

Signature of Authorized Representative*

Anthony.Carna

Date Signed*

07/05/2016

20. PRE-APPLICATION File Name:**21. COVER LETTER ATTACHMENT** File Name:coverletter_final.pdf

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Project/Performance Site Location(s)

Project/Performance Site Primary Location

I am submitting an application as an individual, and not on behalf of a company, state, local or tribal government, academia, or other type of organization.

Organization Name: [REDACTED]
Duns Number: [REDACTED]
[REDACTED] [REDACTED]
Street2:
City*: [REDACTED]
County:
State*: [REDACTED]
Province:
Country*: USA: UNITED STATES
Zip / Postal Code*: [REDACTED]
Project/Performance Site Congressional District*: [REDACTED]

File Name

Additional Location(s)

Project Summary

The brain has the remarkable capability to change in response to experience. This plasticity is essential for learning and memory, and is an important feature of the auditory cortex, especially for learning the significance of sensory signals such as speech, for the use of devices such as cochlear implants, and for recovery after short-term deafness. These changes are thought to occur primarily at synapses, basic units of information processing and plasticity. Long-term synaptic plasticity requires sensory experience and activation of neuromodulatory systems which convey behavioral context to local cortical circuits. However, little is known about the interactions between synaptic inputs and release of neuromodulators in vivo, making it challenging to relate perceptual learning to plasticity in the auditory cortex or other brain areas. Recently we developed an approach to measuring dynamics of synaptic modifications for hours, coupled with imaging techniques enabling us to monitor the same cells over days during training, directly monitoring and manipulating activity in behaving mice. These approaches allow for a close examination of links between modulation, cortical plasticity and auditory perceptual learning.

Specifically, we will study how auditory perceptual training activates the cholinergic vs noradrenergic modulatory systems. These two modulators are principally involved in selective attention towards behaviorally-important stimuli, general arousal, and learning. However, there may be important functional differences in these systems in terms of when they are active during different phases of training or consequences of cholinergic and noradrenergic modulation on auditory neurons for contextual information processing. This proposal describes a series of imaging, recording, optogenetic, and behavioral experiments that will compare and contrast the effects of locus coeruleus activation and norepinephrine release vs the effects of nucleus basalis activation and cholinergic modulation on the primary auditory cortex of behaving mice. Many studies have highlighted the importance of recording in awake animals during behavior, and we will first examine how ensembles of excitatory and inhibitory neurons are affected by learning over the entire duration of training, as animals go from naïve and poor performers, to having reliable performance on an auditory detection and recognition task we have used in the lab for years. Next, we determine when and how cholinergic and noradrenergic modulation affect behavioral and neural responses. Finally, we will make some of the first direct measurements of modulatory neuron responses, asking how these systems are activated by task-relevant variables such as sounds linked to reward.

In summary, here we use in vivo recording and imaging methods to ask how behavioral training engages and modifies noradrenergic and cholinergic systems, to collectively affect auditory cortical processing and persistently improve auditory perceptual abilities in behaving mice.

Project Narrative

Neuroplasticity- the ability of the brain to change in response to experience- is an essential feature of the auditory cortex, especially for speech and language learning as well as the successful use of devices such as cochlear implants. However, it is unclear how motivational state and behavioral training drive plasticity within the central auditory system. The experiments to be performed in this proposal provide essential data on basic mechanisms of neuromodulation and plasticity in the auditory cortex, required for improvement of prosthetic design and therapeutic strategies for treatment of deafness and language disorders.

RESEARCH & RELATED Senior/Key Person Profile (Expanded)

PROFILE - Project Director/Principal Investigator				
Prefix:	First Name*: Robert	Middle Name C	Last Name*: Froemke	Suffix:
Position/Title*:	Assistant Professor			
Organization Name*:	[REDACTED]			
Department:	Otolaryngology			
Division:	[REDACTED]			
Street2:	[REDACTED]			
City*:	[REDACTED]			
Province:	[REDACTED]			
Country*:	USA: UNITED STATES			
Zip / Postal Code*:	[REDACTED]			
Phone Number*:	[REDACTED]	Fax Number:	[REDACTED]	E-Mail*:
Credential, e.g., agency login:	[REDACTED]			
Project Role*:	PD/PI	Other Project Role Category:		
Degree Type:	Ph.D.	Degree Year: 2004		
Attach Biographical Sketch*:	File Name Froemke_Renewal_Biosketch.pdf			
Attach Current & Pending Support:				

B. Positions and Honors

Positions and Employment

2010- Assistant Professor, New York University School of Medicine, Skirball Institute Program in Molecular Neurobiology; Neuroscience Institute; Departments of Otolaryngology, Neuroscience & Physiology; Kimmel Center for Stem Cell Biology; Center for Neural Science.

Other Experience and Professional Memberships

1996-1998 Senior research staff, Center for Connected Learning and Computer-Based Modeling
 2000 Neural Systems & Behavior Course, Marine Biological Laboratory, Woods Hole, MA
 2000- Member, American Association for the Advancement of Science
 2000- Member, Society for Neuroscience
 2004 Grass Fellow, Marine Biological Laboratory, Woods Hole, MA
 2005- Member, Association for Research in Otolaryngology
 2007 Okinawa Computational Neuroscience Course, Okinawa, Japan
 2008 Methods in Computational Neuroscience, Marine Biological Laboratory, Woods Hole, MA
 2009 Teaching Assistant, Biology of Memory Course, Cold Spring Harbor Laboratory, NY
 2010 Kavli Institute for Theoretical Physics, UC Santa Barbara
 2013 Co-Director, Biology & Disorders of Learning & Memory Course, Cold Spring Harbor Lab, NY
 2013-2015 Co-Chair, Cosyne Workshop Committee (2 year appointment)

Honors

1998 Benjamin Brown Prize in Computer Science, Tufts University
 2001 Howard Hughes Medical Institute Predoctoral Fellowship
 2002 Outstanding Graduate Student Instructor, University of California, Berkeley
 2004 First place, General Scientific Meeting presentation, Marine Biological Laboratory
 2005 Jane Coffin Childs Postdoctoral Fellowship
 2006 Sandler Translational Research Postdoctoral Fellowship
 2008 K99/R00 Career Award, NIDCD
 2011 Whitehead Fellowship
 2012 Alfred P. Sloan Research Fellowship Award
 2012 Pew Scholar Award
 2012 Klingenstein Fellowship Award
 2013 NYU Grand Challenge Award
 2013 Hirsch/Weill-Caulier Career Award
 2014 McKnight Scholar Award
 2015 NYU "Next Gen Stars" Inaugural Speaker
 2016 HHMI Faculty Scholars Award

C. Contribution to Science

1. We study the organization and plasticity of cortical synapses, and how cortical modulation and plasticity can improve auditory perception and behavior, in the context of behavioral training and perceptual learning. We have a particular emphasis on conducting well-controlled and parametric studies of auditory psychophysics, combined with mechanistic studies of neuromodulation and long-term plasticity of excitatory and inhibitory synapses. We have conducted a series of studies examining how manipulations of modulatory systems- the cholinergic attentional system of the nucleus basalis and the noradrenergic arousal system of the locus coeruleus - lead to synaptic plasticity and produce behavioral changes in adult rats. We assessed baseline auditory abilities to determine which stimuli were difficult to perceive, and leveraged the cholinergic system to boost up the strengths of synapses at these thresholds. We found that auditory perception was improved for at least hours afterwards, indicating that direct cortical modifications can be useful for enhancing sensory perception and behavior. We have contrasted these changes with the action of noradrenalin and stimulation (electrical or optogenetic) of the rat locus coeruleus, identifying how cholinergic and noradrenergic modulation differentially affect plasticity and perception. This is some of the first work identifying inhibitory synapses and circuits as central targets of neuromodulators, and our findings that acetylcholine disinhibits auditory cortex has been replicated by several other labs, in rats and in mice, and other sensory systems.

- a. **Froemke RC**, Merzenich MM, Schreiner, CE. A synaptic memory trace for cortical receptive field plasticity. **Nature** 2007; 450:425-429. PMID: 18004384 PMC in process
 - b. **Froemke RC**, Carcea I, Barker AJ, Yuan K, Seybold B, Martins ARO, Zaika N, Bernstein H, Wachs M, Levis PA, Polley DB, Merzenich MM, Schreiner CE. Long-term modification of cortical synapses improves sensory perception. **Nature Neuroscience** 2013; 16:79-88. PMC: 3711827
 - c. **Froemke RC**. Plasticity of cortical excitatory-inhibitory balance. **Annual Review of Neuroscience** 2015; 38:195-219. PMID:25897875 PMC:4652600
 - d. Martins ARO, **Froemke RC**. Coordinated forms of noradrenergic plasticity in the locus coeruleus and primary auditory cortex. **Nature Neuroscience** 2015; 18:1483-1492. PMID:26301326 PMC: 4583810
2. Excitatory-inhibitory balance is an important property of mature neural circuits, ensuring that excitability is carefully controlled for information processing without seizure generation or propagation failure. How are inhibitory inputs calibrated during development and adjusted throughout life, to ensure that inhibition balances excitation? Inhibitory maturation is believed to determine critical periods for cortical development, and changes to excitatory synapses related to learning must be matched by coordinated changes in co-tuned inhibitory inputs. Our lab specializes in examining combined forms of excitatory and inhibitory long-term synaptic plasticity with whole-cell recordings and 2-photon imaging in vivo, monitoring the dynamics by which excitation and inhibition are adjusted after changes in patterns of electrical activity or sensory experience. We have shown how these processes occur during a critical period for frequency tuning in the rodent auditory cortex. We are also the first and perhaps only group to show how, after transplantation, inhibitory progenitor cells integrate into the existing network to open a new critical period in adult visual cortex, using paired recordings from host and transplanted cells to show that these new neurons make and receive synaptic connections in the local circuit.
- a. Dornn A, Yuan K, Barker AJ, Schreiner CE, **Froemke RC**. Developmental sensory experience balances cortical excitation and inhibition. **Nature** 2010; 465:932-936. PMID:20559387 PMC2888507
 - b. Southwell DG, **Froemke RC**, Alvarez-Buylla A, Stryker MP, Gandhi SP. Cortical plasticity induced by inhibitory neuron transplantation. **Science** 2010; 327:1145-1148. PMID:20185728 PMC:3164148
 - c. Southwell DG, Paredes MF, Galvao RP, Jones DL, **Froemke RC**, Sebe JY, Alfaro-Cervello C, Garcia-Verdugo JM, Baraban SC, Alvarez-Buylla A. Intrinsically determined cell death of developing cortical interneurons. **Nature** 2012; 491:109-113. PMID:23041929 PMC:3726009
 - d. Cohen S, Ma H, Kuchibhotla K, Watson BO, Buzsáki G, **Froemke RC**, Tsien RW. Excitation-transcription coupling in parvalbumin-positive interneurons employs a novel CaM Kinase-dependent pathway distinct from excitatory neurons. **Neuron** 2016; 90:292-307. PMID:27041500 PMC:4866871
3. Our studies of neuromodulation and behavior also examine circuit dynamics and the control of social cognition, with a particular emphasis on oxytocin. It has historically been difficult to determine how modifications of specific synapses relate to changes in behavior. We have examined how neurons in the rodent hypothalamus affect synaptic transmission in the cortex and elsewhere to produce behavioral changes in adult rats and mice. This is some of the first work using cortical plasticity to persistently enhance sensory perception and cognition. Furthermore, we provided the first direct evidence that oxytocin transiently reduces synaptic inhibition in the cortex, increasing the salience of incoming sensory inputs. We have used optogenetics and pharmacological approaches to examine how oxytocin can enable newly-maternal mice to recognize the significance of infant vocalizations and distress calls. As part of our work on oxytocin, in collaboration with Moses Chao's lab we generated the first specific antibodies to the mouse oxytocin receptor, which we have shared with many international laboratories (including the Grinevich and Stoop labs). We have also worked to understand the functional anatomy and circuit logic by which oxytocin neurons and other hypothalamic cell types project to target areas.
- a. Marlin BJ, Mitre M, D'amour JA, Chao MV, **Froemke RC**. Oxytocin enables maternal behaviour by balancing cortical inhibition. **Nature** 2015; 520:499-504. PMID: 25874674 PMC: 4409554
 - b. Mitre M, Marlin BJ, Schiavo JK, Morina E, Norden S, Hackett TA, Aoki C, Chao MV, **Froemke RC**. A distributed network for social cognition enriched for oxytocin receptors. **Journal of Neuroscience** 2016; 36:2517-2535. PMID: 26911697 PMC: 4764667
 - c. Eliava M, Melchior M, Knobloch-Bollmann S, Wahis J, da Silva Gouveia M, Tang Y, Ciobanu AC, del Rio RT, Roth LC, Althammer F, Chavant V, Goumon Y, Gruber T, Petit-Demoulière M, Busnelli M, Chini B, Tan L, Mitre M, **Froemke RC**, Chao MV, Giese G, Sprengel R, Kuner R, Poisbeau P, Seeburg PH, Stoop R, Charlet A, Grinevich V. A new population of parvocellular oxytocin neurons controlling

magnocellular neuron activity and inflammatory pain processing. **Neuron** 2016; 89:1291-1304. PMID: 26948889

- d. Wong LC, Wang L, Yumita T, D'amour JA, Chen G, Chang B, Bernstein H, You X, Feng J, **Froemke RC**, Lin D. Effective modulation of male aggression through the lateral septum to medial hypothalamus projection. **Current Biology** 2016; 26:593-604. PMID: 26877081 PMC: 4783202
4. Neural activity can be complex. We have examined spike-timing-dependent plasticity (STDP), focusing on synaptic modifications induced by naturalistic patterns of pre- and postsynaptic spikes recorded in vivo. From hundreds of experiments we could predict the sign and magnitude of long-term synaptic plasticity induced by complex spike trains, and continue to work with computational neuroscientists to understand how STDP might enable neural networks to store and recall information. Some experiments examine how synaptic integration and dendritic properties affect NMDA receptor activation to control induction of synaptic plasticity. More recently, we are examining how multiple synapses are co-modified, and we were the first group to show that excitatory and inhibitory STDP can be coordinated and induced together. We found that spike pairing can normalize the strength of inhibition relative to the strength of co-activated excitation, providing a natural mechanism by which excitatory-inhibitory balance can be established and maintained.
 - a. **Froemke RC**, Dan Y. Spike-timing-dependent synaptic modification induced by natural spike trains. **Nature** 2002; 416:433-438. PMID: 11919633
 - b. **Froemke RC**, Poo MM, Dan Y. Spike-timing-dependent plasticity depends on dendritic location. **Nature** 2005; 434:221-225. PMID: 15759002
 - c. Ponte Costa R, **Froemke RC**, Sjöström PJ, van Rossum MC. Unified pre- and postsynaptic long-term plasticity enables reliable and flexible learning. **eLife** 2015; 4. PMID: 26308579 PMC:4584257
 - d. D'amour JA, **Froemke RC**. Inhibitory and excitatory spike-timing-dependent plasticity in the auditory cortex. **Neuron** 2015; 86:514-528. PMID: 25843405 PMC: 4409545
 5. Studies of plasticity in the auditory cortex have the potential to inform and transform training procedures and use of neuroprosthetic devices such as cochlear implants. Our lab has pioneered a new rat model of multi-channel cochlear implant use, behaviorally and physiologically validated. This is being combined with μ -ECoG recordings for stable, long-term monitoring of cortical changes with use of the implants.
 - a. Wang J, Trumpis M, Insanally M, **Froemke R**, Viventi J. A low-cost, multiplexed electrophysiology system for chronic μ ECoG in rodents. Conf Proc IEEE Eng Med Biol Soc 2014; 2014:5256-5259. PMID: 25571179 PMC: in process
 - b. King J, Insanally M, Jin M, Martins ARO, D'amour JA, **Froemke RC**. Rodent auditory perception: critical band limitations and plasticity. **Neuroscience** 2015; 296:55-65. PMID: 27281743
 - c. Insanally M, Trumpis M, Wang C, Chiang CH, Woods V, Bossi S, **Froemke RC**, Viventi J. A low-cost, multiplexed μ ECoG system for long-term, reliable high-density recordings in rodents. **Journal of Neural Engineering** 2016; 13:026030. PMID: 26975462, PMCID: in process.
 - d. King J, Shehu I, Roland JT, Svirsky MA, **Froemke RC**. A physiological and behavioral system for hearing restoration with cochlear implants. **Journal of Neurophysiology** 2016; in press.

Complete List of Published Work in MyBibliography:

<http://www.ncbi.nlm.nih.gov/sites/myncbi/robert.froemke.1/bibliography/44917017/public/?sort=date&direction=ascending>

D. Research Support

Ongoing Research Support

HHMI Faculty Scholars Award

2016-2021

Cortical plasticity and control of social behavior

This career award supports our research on the mammalian oxytocin system, and cortical plasticity for learned maternal behavior. Role: PI

NYU CTSI Collaborative Translational Pilot Project Program

2016-2017

Optimizing cochlear implant use with neural recordings

The goal of this study is to understand the neural basis of cochlear implant use in trained, deafened rats. Role: Co-PI (with Mario Svirsky, NYU)

- NYU Global Seed Grant for Collaborative Research 2016-2018
 Neuromodulatory influences on economic preferences
 The goal of this study is to examine the neurobiological mechanisms for impulsivity behaviors in trained rats.
 Role: Co-PI (with Jeff Erlich, NYU Shanghai)
- Simons Foundation Explorer Grant 2016-2017
 Synthetic infrared nanosensors for real-time monitoring of oxytocin release
 The goal of this study is to screen carbon nanotube-based sensors for specific detection of oxytocin in real time. Role: Co-PI (with Markita Landry, UC Berkeley)
- McKnight Scholar Award 2014-2017
 Neural circuitry and plasticity for control of mammalian social behavior
 The goal of this study is to determine the 'social receptive field' of oxytocin neurons in the hypothalamus, and relate activation of the oxytocin system and cortical plasticity to learned maternal behavior. Role: PI
- Hirsch/Weill-Caulier Career Award 2014-2018
 Neural basis of long-term memory formation
 The goal of this study is to examine how neuromodulatory circuits of the locus coeruleus are modified by sensory experience and neural activity. Role: PI
- R01 NIDCD DC012557 2012-2017
 Synaptic basis for perceptual learning in primary auditory cortex
 The goal of this study is to directly examine the relation between adult cortical synaptic plasticity and perceptual learning via the noradrenergic modulatory system. Role: PI
- Completed Research Support**
- K99/R00 NIDCD DC009635 2008-2015
 Synaptic basis for perceptual learning in primary auditory cortex
 The goal of this study was to directly examine the relation between adult cortical synaptic plasticity and perceptual learning via the cholinergic modulatory system. Role: PI
- Pew Scholarship 2012-2016
 Neural basis of learned social behavior
 The goal of this study was to determine how oxytocin affects responses to pup calls in the auditory cortex. Role: PI
- Klingenstein Fellowship 2012-2015
 Plasticity of excitatory-inhibitory balance in the auditory cortex
 The goal of this study was to determine the network dynamics and mechanisms that calibrate and balance excitation and inhibition in the developing auditory cortex. Role: PI
- Alfred P. Sloan Research Fellowship 2012-2014
 Synaptic plasticity in the cerebral cortex
 The goal of this study was to examine the functional consequences of long-term synaptic plasticity in cortical networks for behavioral modification. Role: PI
- NYU Provost's Grand Challenge Grant 2013-2015
 Smart neuroprosthetics: brain-machine interfaces for the 21st century
 The goal of this study was to generate brain-machine interface devices for improving sensory, motor, and cognitive behavioral performance in rodents and primates. Role: Co-PI (with Michael Long, Bijan Pesaran, Dan Sanes, and Jonathan Viventi, NYU)
- NYU Provost's Office Mega-Grants Initiative 2013-2014
 Creating a neuroengineering core facility at NYU
 The goal of this study was to seed construction of a core facility at NYU for fabrication of neuroprosthetic devices. Role: Co-PI (with Michael Long, Dan Sanes, John Simson, and Jonathan Viventi, NYU)

RESEARCH & RELATED BUDGET - SECTION A & B, Budget Period 1

ORGANIZATIONAL DUNS*: [REDACTED]

Budget Type*: Project Subaward/Consortium

Enter name of Organization: [REDACTED]

Start Date*: 04-01-2017 End Date*: 03-31-2018 Budget Period: 1

A. Senior/Key Person													
Prefix	First Name*	Middle Name	Last Name*	Suffix	Project Role*	Base Salary (\$)	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits (\$)*	Funds Requested (\$)*	
1.	Robert	C	Froemke		PD/PI	[REDACTED]	5	0	0	[REDACTED]	[REDACTED]	[REDACTED]	
Total Funds Requested for all Senior Key Persons in the attached file													
											[REDACTED]		
Additional Senior Key Persons:		File Name:									Total Senior/Key Person		[REDACTED]

B. Other Personnel											
Number of Personnel*	Project Role*	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits*	Funds Requested (\$)*				
2	Post Doctoral Associates	15	0	0	[REDACTED]	[REDACTED]	[REDACTED]				
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]				
	Undergraduate Students										
	Secretarial/Clerical										
1	Technician	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]				
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]				
Total Salary, Wages and Fringe Benefits (A+B)							[REDACTED]				

RESEARCH & RELATED Budget {A-B} (Funds Requested)

RESEARCH & RELATED BUDGET - SECTION A & B, Budget Period 2

ORGANIZATIONAL DUNS*: [REDACTED]

Budget Type*: Project Subaward/Consortium

Enter name of Organization: [REDACTED]

Start Date*: 04-01-2018

End Date*: 03-31-2019

Budget Period: 2

A. Senior/Key Person												
Prefix	First Name*	Middle Name	Last Name*	Suffix	Project Role*	Base Salary (\$)	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits (\$)*	Funds Requested (\$)*
1.	Robert	C	Froemke		PD/PI	[REDACTED]	5	0	0	[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]												[REDACTED]
[REDACTED]												[REDACTED]

B. Other Personnel									
Number of Personnel*	Project Role*	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits*	Funds Requested (\$)*		
1	Post Doctoral Associates	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]
	Undergraduate Students								
	Secretarial/Clerical								
1	Technician	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]

RESEARCH & RELATED Budget {A-B} (Funds Requested)

RESEARCH & RELATED BUDGET - SECTION C, D, & E, Budget Period 2

ORGANIZATIONAL DUNS*: [REDACTED]

Budget Type*: Project Subaward/Consortium

Organization: [REDACTED]

Start Date*: 04-01-2018

End Date*: 03-31-2019

Budget Period: 2

C. Equipment Description		Funds Requested (\$)*
List items and dollar amount for each item exceeding \$5,000		
Equipment Item		
Total funds requested for all equipment listed in the attached file		[REDACTED]
Total Equipment		[REDACTED]
Additional Equipment: File Name:		

D. Travel	Funds Requested (\$)*
1. Domestic Travel Costs (Incl. Canada, Mexico, and U.S. Possessions)	[REDACTED]
[REDACTED]	[REDACTED]
	[REDACTED]

E. Participant/Trainee Support Costs	Funds Requested (\$)*
1. Tuition/Fees/Health Insurance	
2. Stipends	
3. Travel	
4. Subsistence	
5. Other:	
Number of Participants/Trainees	Total Participant Trainee Support Costs

RESEARCH & RELATED Budget (C-E) (Funds Requested)

RESEARCH & RELATED BUDGET - SECTIONS F-K, Budget Period 2

ORGANIZATIONAL DUNS*: [REDACTED]
 Budget Type*: Project Subaward/Consortium
 Organization: [REDACTED]

Start Date*: 04-01-2018 End Date*: 03-31-2019 Budget Period: 2

F. Other Direct Costs	Funds Requested (\$)*
1. Materials and Supplies	
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
Total	[REDACTED]

G. Direct Costs	Funds Requested (\$)*
Total Direct Costs (A thru F)	[REDACTED]

H. Indirect Costs			
Indirect Cost Type	Indirect Cost Rate (%)	Indirect Cost Base (\$)	Funds Requested (\$)*
1. Research (MTDC)	[REDACTED]	[REDACTED]	[REDACTED]
		[REDACTED]	[REDACTED]
Cognizant Federal Agency	DHHS, [REDACTED]		
<small>(Agency Name, POC Name, and POC Phone Number)</small>			

I. Total Direct and Indirect Costs	Funds Requested (\$)*
Total Direct and Indirect Institutional Costs (G + H)	[REDACTED]

J. Fee	Funds Requested (\$)*
	[REDACTED]

K. Budget Justification*	File Name: budget_justification_final.pdf (Only attach one file.)
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RESEARCH & RELATED Budget (F-K) (Funds Requested)

RESEARCH & RELATED BUDGET - SECTION A & B, Budget Period 3

ORGANIZATIONAL DUNS*: [REDACTED]

Budget Type*: Project Subaward/Consortium

Enter name of Organization: [REDACTED]

Start Date*: 04-01-2019 End Date*: 03-31-2020 Budget Period: 3

A. Senior/Key Person												
Prefix	First Name*	Middle Name	Last Name*	Suffix	Project Role*	Base Salary (\$)	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits (\$)*	Funds Requested (\$)*
1.	Robert	C	Froemke		PD/PI	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]												[REDACTED]
[REDACTED]												[REDACTED]

B. Other Personnel									
Number of Personnel*	Project Role*	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits*	Funds Requested (\$)*		
1	Post Doctoral Associates	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]		
	Undergraduate Students								
	Secretarial/Clerical								
1	Technician	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]		

RESEARCH & RELATED Budget {A-B} (Funds Requested)

RESEARCH & RELATED BUDGET - SECTION C, D, & E, Budget Period 3

ORGANIZATIONAL DUNS*: [REDACTED]

Budget Type*: Project Subaward/Consortium

Organization: [REDACTED]

Start Date*: 04-01-2019

End Date*: 03-31-2020

Budget Period: 3

C. Equipment Description		Funds Requested (\$)*
List items and dollar amount for each item exceeding \$5,000		
Equipment Item		
Total funds requested for all equipment listed in the attached file		[REDACTED]
[REDACTED]		[REDACTED]
Additional Equipment: File Name:		

D. Travel		Funds Requested (\$)*
1. Domestic Travel Costs (Incl. Canada, Mexico, and U.S. Possessions)		
[REDACTED]		[REDACTED]
[REDACTED]		[REDACTED]

E. Participant/Trainee Support Costs		Funds Requested (\$)*
1. Tuition/Fees/Health Insurance		
2. Stipends		
3. Travel		
4. Subsistence		
5. Other:		
Number of Participants/Trainees	Total Participant Trainee Support Costs	[REDACTED]

RESEARCH & RELATED Budget (C-E) (Funds Requested)

RESEARCH & RELATED BUDGET - SECTIONS F-K, Budget Period 3

ORGANIZATIONAL DUNS*: [REDACTED]
 Budget Type*: Project Subaward/Consortium
 Organization: [REDACTED]

Start Date*: 04-01-2019 End Date*: 03-31-2020 Budget Period: 3

F. Other Direct Costs	Funds Requested (\$)*
1. Materials and Supplies	
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
Total	[REDACTED]

G. Direct Costs	Funds Requested (\$)*
Total Direct Costs (A thru F)	[REDACTED]

H. Indirect Costs			
Indirect Cost Type	Indirect Cost Rate (%)	Indirect Cost Base (\$)	Funds Requested (\$)*
1. Research (MTDC)	[REDACTED]	[REDACTED]	[REDACTED]
		[REDACTED]	[REDACTED]
Cognizant Federal Agency	DHHS, [REDACTED]		
<small>(Agency Name, POC Name, and POC Phone Number)</small>			

I. Total Direct and Indirect Costs	Funds Requested (\$)*
Total Direct and Indirect Institutional Costs (G + H)	[REDACTED]

J. Fee	Funds Requested (\$)*
	[REDACTED]

K. Budget Justification*	File Name: budget_justification_final.pdf (Only attach one file.)
---------------------------------	--

RESEARCH & RELATED Budget (F-K) (Funds Requested)

RESEARCH & RELATED BUDGET - SECTION A & B, Budget Period 4

ORGANIZATIONAL DUNS*: [REDACTED]

Budget Type*: Project Subaward/Consortium

Enter name of Organization: [REDACTED]

Start Date*: 04-01-2020 End Date*: 03-31-2021 Budget Period: 4

A. Senior/Key Person												
Prefix	First Name*	Middle Name	Last Name*	Suffix	Project Role*	Base Salary (\$)	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits (\$)*	Funds Requested (\$)*
1.	Robert	C	Froemke		PD/PI	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]												[REDACTED]
[REDACTED]												[REDACTED]

B. Other Personnel									
Number of Personnel*	Project Role*	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits*	Funds Requested (\$)*		
1	Post Doctoral Associates	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]		
	Undergraduate Students								
	Secretarial/Clerical								
1	Technician	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]		

RESEARCH & RELATED Budget {A-B} (Funds Requested)

RESEARCH & RELATED BUDGET - SECTION A & B, Budget Period 5

ORGANIZATIONAL DUNS*: [REDACTED]

Budget Type*: Project Subaward/Consortium

Enter name of Organization: [REDACTED]

Start Date*: 04-01-2021 End Date*: 03-31-2022 Budget Period: 5

A. Senior/Key Person												
Prefix	First Name*	Middle Name	Last Name*	Suffix	Project Role*	Base Salary (\$)	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits (\$)*	Funds Requested (\$)*
1.	Robert	C	Froemke		PD/PI	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]												[REDACTED]
[REDACTED]												[REDACTED]

B. Other Personnel									
Number of Personnel*	Project Role*	Calendar Months	Academic Months	Summer Months	Requested Salary (\$)*	Fringe Benefits*	Funds Requested (\$)*		
1	Post Doctoral Associates	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]		
	Undergraduate Students								
	Secretarial/Clerical								
1	Technician	12	0	0	[REDACTED]	[REDACTED]	[REDACTED]		
[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]	[REDACTED]		

RESEARCH & RELATED Budget {A-B} (Funds Requested)

RESEARCH & RELATED BUDGET - SECTION C, D, & E, Budget Period 5

ORGANIZATIONAL DUNS*: 121911077

Budget Type*: Project Subaward/Consortium

Organization: New York University School Of Medicine

Start Date*: 04-01-2021

End Date*: 03-31-2022

Budget Period: 5

C. Equipment Description		Funds Requested (\$)*
List items and dollar amount for each item exceeding \$5,000		
Equipment Item		
Total funds requested for all equipment listed in the attached file		[REDACTED]
	[REDACTED]	[REDACTED]
Additional Equipment: File Name:		

D. Travel	Funds Requested (\$)*
1. Domestic Travel Costs (Incl. Canada, Mexico, and U.S. Possessions)	[REDACTED]
[REDACTED]	[REDACTED]
	[REDACTED]

E. Participant/Trainee Support Costs	Funds Requested (\$)*
1. Tuition/Fees/Health Insurance	
2. Stipends	
3. Travel	
4. Subsistence	
5. Other:	
Number of Participants/Trainees	Total Participant Trainee Support Costs

RESEARCH & RELATED Budget (C-E) (Funds Requested)

RESEARCH & RELATED BUDGET - SECTIONS F-K, Budget Period 5

ORGANIZATIONAL DUNS*: [REDACTED]
 Budget Type*: Project Subaward/Consortium
 Organization: [REDACTED]

Start Date*: 04-01-2021 End Date*: 03-31-2022 Budget Period: 5

F. Other Direct Costs	Funds Requested (\$)*
1. Materials and Supplies	
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]
Total	[REDACTED]

G. Direct Costs	Funds Requested (\$)*
Total Direct Costs (A thru F)	[REDACTED]

H. Indirect Costs			
Indirect Cost Type	Indirect Cost Rate (%)	Indirect Cost Base (\$)	Funds Requested (\$)*
1. Research (MTDC)	[REDACTED]	[REDACTED]	[REDACTED]
		[REDACTED]	[REDACTED]
Cognizant Federal Agency		DHHS, Darryl W. Mayes 212-264-2069	
<small>(Agency Name, POC Name, and POC Phone Number)</small>			

I. Total Direct and Indirect Costs	Funds Requested (\$)*
Total Direct and Indirect Institutional Costs (G + H)	[REDACTED]

J. Fee	Funds Requested (\$)*
	[REDACTED]

K. Budget Justification*	File Name: budget_justification_final.pdf (Only attach one file.)
---------------------------------	--

RESEARCH & RELATED Budget (F-K) (Funds Requested)

Travel

We are requesting funds to support the PI's travel to two meetings each year (the Society for Neuroscience annual meeting and either Cosyne or the Association for Research in Otolaryngology midwinter meeting), as well as support the attendance of the postdocs and graduate students at one meeting each year.

RESEARCH & RELATED BUDGET - Cumulative Budget

	Totals (\$)	
Section A, Senior/Key Person		██████████
██████████		██████████
Total Number Other Personnel	16	
Total Salary, Wages and Fringe Benefits (A+B)		██████████
██████████		██████████
██████████		██████████
1. Domestic	██████████	
██████████	██████████	
Section E, Participant/Trainee Support Costs		0.00
1. Tuition/Fees/Health Insurance	0.00	
2. Stipends	0.00	
3. Travel	0.00	
4. Subsistence	0.00	
5. Other	0.00	
6. Number of Participants/Trainees	0	
Section F, Other Direct Costs		██████████
1. Materials and Supplies	██████████	
2. Publication Costs	0.00	
3. Consultant Services	0.00	
4. ADP/Computer Services	0.00	
5. Subawards/Consortium/Contractual Costs	0.00	
6. Equipment or Facility Rental/User Fees	██████████	
██████████	██████████	
8. Other 1	0.00	
9. Other 2	0.00	
10. Other 3	0.00	
Section G, Direct Costs (A thru F)		██████████
██████████		██████████
██████████		██████████
██████████		██████████

PHS 398 Cover Page Supplement

4. Human Embryonic Stem Cells Section

*Does the proposed project involve human embryonic stem cells? Yes No

If the proposed project involves human embryonic stem cells, list below the registration number of the specific cell line(s) from the following list: http://grants.nih.gov/stem_cells/registry/current.htm. Or, if a specific stem cell line cannot be referenced at this time, please check the box indicating that one from the registry will be used:

Specific stem cell line cannot be referenced at this time. One from the registry will be used.

Cell Line(s) (Example: 0004):

5. Inventions and Patents Section (RENEWAL)

*Inventions and Patents: Yes No

If the answer is "Yes" then please answer the following:

*Previously Reported: Yes No

6. Change of Investigator / Change of Institution Section

Change of Project Director / Principal Investigator

Name of former Project Director / Principal Investigator

Prefix:

*First Name:

Middle Name:

*Last Name:

Suffix:

Change of Grantee Institution

*Name of former institution:

PHS 398 Research Plan

OMB Number: 0925-0001

Expiration Date: 10/31/2018

Introduction

1. Introduction to Application

(Resubmission and Revision)

Research Plan Section

2. Specific Aims

aims_final.pdf

3. Research Strategy*

research_strategy_final.pdf

4. Progress Report Publication List

progress_report_publication_list.pdf

Human Subjects Section

5. Protection of Human Subjects

6. Data Safety Monitoring Plan

7. Inclusion of Women and Minorities

8. Inclusion of Children

Other Research Plan Section

9. Vertebrate Animals

animals_final.pdf

10. Select Agent Research

11. Multiple PD/PI Leadership Plan

12. Consortium/Contractual Arrangements

13. Letters of Support

support_letters_final2.pdf

14. Resource Sharing Plan(s)

resource_sharing_final.pdf

15. Authentication of Key Biological and/or Chemical Resources

resource_authentication_final.pdf

Appendix

16. Appendix

I. Specific Aims

Synaptic basis of perceptual learning in primary auditory cortex

The goal of this project is to determine how cholinergic and noradrenergic modulatory systems collectively promote synaptic plasticity in primary auditory cortex (AI), to improve auditory perceptual abilities in behaving mice. Modulation and plasticity are important features of AI, especially for forming representations of sensory signals such as speech, music, or other behaviorally-relevant sounds¹⁻¹⁰. Changes in neural circuits and behavior can be incredibly long-lasting after auditory conditioning¹¹⁻¹⁴, but the mechanisms by which AI networks are persistently modified and affect auditory perception are unclear. In particular, it remains challenging to connect perceptual learning to plasticity in AI or elsewhere in the brain. Impaired cortical processing and plasticity are believed to occur in disorders such as hearing loss, language impairments, and tinnitus¹⁵⁻¹⁷; conversely, enabling plasticity by training programs and devices such as hearing aids and cochlear implants may improve outcomes in pathological conditions and hearing loss¹⁸⁻²². Successful completion of this project will result in a significantly improved mechanistic model of auditory learning and AI plasticity, critical for studies of language learning, speech processing, deafness, and the design of medical devices and training procedures for hearing restoration.

Adult cortical plasticity requires sensory experience and neuromodulation, which relays global behavioral context to local cortical circuits. Many modulatory systems, including the noradrenergic system of the locus coeruleus²³⁻³⁰ and the cholinergic system of the basal forebrain^{1,2,31-40}, are recruited by surprising or arousing stimuli, and promote plasticity in target circuits including rodent AI. What are the functional differences between acetylcholine and norepinephrine for perceptual learning and auditory behavior? Specifically, here we will ask how training affects AI circuits during behavior (Aim 1); if cholinergic and noradrenergic systems are recruited at distinct times, to differentially modulate AI during auditory training for enhanced perception (Aim 2); and finally, when and how neuromodulatory neurons become responsive to task-relevant sounds and other cues during training (Aim 3).

Previously we studied cholinergic or noradrenergic modulation separately in behaving rats, measuring physiological changes under anesthesia^{10,28,35,36}. Here we will relate behavior to neural activity with in vivo whole-cell recording and 2-photon imaging from AI and modulatory neurons in behaving head-fixed mice. These studies take advantage of our past work on AI modulation and plasticity^{28,35,36,41-45}, using a novel approach we developed to document synaptic plasticity in multiple cells over days, in behaving mice performing an auditory task. This allows us to measure and manipulate modulatory systems on a trial-by-trial basis while monitoring responses over the entire time-course of learning in individual mice. We hypothesize that initially, norepinephrine reduces tonic inhibition to increase excitability to all stimuli; this may increase detection and behavioral responses but at the cost of false alarms. As mice learn the task over days, acetylcholine then selectively enhances a sub-population of excitatory and inhibitory AI neurons for reliable task performance. Our goal is a unified model of cholinergic and noradrenergic modulation for AI excitatory and inhibitory plasticity important for behavior.

Aim 1. To determine how AI excitatory/inhibitory cells and synapses are modified by auditory training

We will train adult mice on a go/no-go auditory task, to examine perceptual learning by measuring detection thresholds and sound recognition abilities. We will use in vivo 2-photon imaging (Subaim 1a) and whole-cell recordings (Subaim 1b) from AI neurons in behaving mice to determine how excitatory/inhibitory inputs and outputs are modified by learning, and assess if these changes are required for task performance.

Aim 2. To ask how cholinergic and noradrenergic modulation affect auditory learning and AI plasticity

We will record from AI during behavior, using optogenetics and pharmacology to ask if and when the cholinergic vs noradrenergic systems are necessary for initial learning and maintained peak performance (Subaim 2a). We will ask if pairing task-relevant sounds with cholinergic and/or noradrenergic modulation can accelerate learning or enhance peak performance (Subaim 2b). We will determine the synaptic mechanisms by which cholinergic and noradrenergic modulation enable AI excitatory/inhibitory plasticity during learning.

Aim 3. To test the hypothesis that noradrenergic and cholinergic neurons respond to task variables

How are the noradrenergic and cholinergic systems recruited when mice are engaged in the task? We will ask if there is a sequence of AI modulation over task learning; specifically if the noradrenergic system is recruited transiently during the first day, while the cholinergic system comes on-line over subsequent days and is needed for steady-state performance. We will record from locus coeruleus and nucleus basalis, and perform 2-photon imaging of modulatory axons within AI in trained animals and over the duration of perceptual learning. We also recently found that tone-evoked responses with short latency are induced in locus coeruleus neurons after pairing tones with locus coeruleus stimulation²⁸. Here we will ask if plasticity in nucleus basalis and/or locus coeruleus is important for auditory learning and task performance.

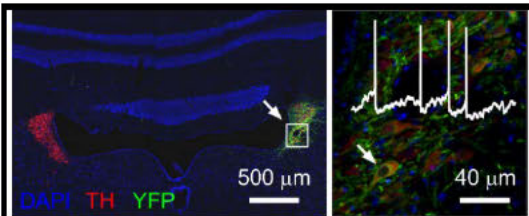


Figure 15. Immunohistochemistry: LC whole-cell recording and biocytin fill.

determine cell identity (Fig. 15). While it will be challenging to find responsive nucleus basalis cells, this is a ‘high-risk, high-reward’ experiment, not critical to other goals of this project, and even a few cells (~4-5 required for statistical power) may be informative. We are routinely able to obtain ~5-10 recordings per session, including cell-attached recordings, allowing us to screen many cells.

5) Finally, we aim to determine if these changes in modulatory neural responses are due to local plasticity in subcortical areas, and if preventing this plasticity affects learning. Modulatory neurons are activated by task context, and develop responses to conditioned stimuli after training^{23,24,28}. Emergence of auditory responses in nucleus basalis or locus coeruleus (with short latency and present even under anesthesia; Fig. 5) are potentially essential components by which previously-innocuous stimuli develop meaning, and thus it is critical to understand the mechanisms and behavioral relevance of these changes for auditory learning. Here we perform two studies, asking first if infusion of APV (to block NMDA receptors) or modulatory receptor antagonists prevent auditory responses or responses to context cues (e.g., licktube) from arising in these nuclei. We will then infuse these blockers into either locus coeruleus or nucleus basalis on the first day of training (in some animals) or each day of training (in other animals) to determine how behavioral performance and learning is impacted when modulatory plasticity is blocked.

Discussion/Alternatives: We aim to obtain a multi-dimensional view of how behavioral context and task variables differentially engage cholinergic vs. noradrenergic modulatory systems. This entails examining subthreshold vs suprathreshold responses, and local vs more global signals over training. We hypothesize that the presence of the licktube (which signals active behavioral context) initially activates locus coeruleus, before these neurons begin responding to task-relevant sounds (both targets and foils; Fig. 14d,g). As performance improves, locus coeruleus activation sharply declines, and instead cholinergic neurons begin responding (shown by our preliminary data in Fig. 14a-c). We will examine the behavioral importance of the emergence of these responses. Our past work (Fig. 5) showed that NMDA receptors were critical for this plasticity, but we will examine other candidate mechanisms (e.g., modulatory autoreceptors) if necessary. In future studies beyond the scope here, we will map the functional anatomy that provides potential auditory information to modulatory centers.

These are technically-challenging methods, but we have experience and success with each approach. Past studies showed that surprising events or contextual cues increase cholinergic or noradrenergic tone, often measured with amperometry in rodent and primate cortex^{64,65,150}. However, this is indirect with low spatiotemporal resolution, making it difficult to relate degree of modulatory activation with single-trial neural or behavioral responses. Instead, here we take advantage of mouse lines for modulatory control, for optical identification of cell-types (Fig. 14). This is important, as locus coeruleus to some degree and nucleus basalis to a potentially high degree are neurochemically heterogeneous^{29,39,65}. In vivo whole-cell recordings might reveal if even in naïve (untrained) animals, these neurons receive sensory-evoked synaptic inputs, but these inputs might be subthreshold or masked by inhibition to keep modulatory cells from spiking. It may be technically infeasible to make in vivo whole-cell recordings from cholinergic cells given their sparsity; we and others have already made in vivo whole-cell recordings from locus coeruleus noradrenergic neurons^{28,151}. Conversely, it may be difficult to perform tetrode recording from locus coeruleus. For these reasons we use multiple methods to ask the same questions, and provide new data on spatial and temporal patterns of AI neuromodulation during auditory training.

Summary: Neuromodulation is important for auditory plasticity and perceptual learning. However, it is unclear how two of the main modulators involved in these processes- acetylcholine and norepinephrine- are naturally recruited during auditory training to differentially modulate target circuits. Our studies will provide new insight into these mechanisms, reveal how cortical plasticity is involved in auditory behavior, and suggest how modulatory systems are activated and modified by training for effective control of auditory perception. In Aim 1, we ask how auditory training relates to AI plasticity measured in behaving mice. We perform *in vivo* whole-cell recordings and imaging to determine dynamics of response changes. In Aim 2, we examine distinctions between cholinergic and noradrenergic modulation at different phases of learning. In Aim 3, we record from these systems and ask if they are plastic, relating auditory responses in modulatory axons and neurons to auditory perceptual learning.

AIMS	YR 1	YR 2	YR 3	YR 4	YR 5
1a. AI imaging during behavior	KK,EM,RCF				
1b. In vivo whole-cell recordings	KK,NL,RCF				
2a. Necessity of AI neuromod	KK,NL,RCF				
2b. Does AI mod aid learning?		KK,NL			
3. Plasticity of neuromodulation		KK,NL, EM,RCF			

Feasibility: All Aims are feasible in 5 yrs.

OR successors thereafter.

Progress Report Publication List

For the original R01 proposal, we published several papers related to rodent auditory perception and cortical plasticity, including one major study (Martins and Froemke, *Nature Neuroscience* 2015) that constitutes Aim 1a, 1b, and almost all of the proposed studies in Aims 2 and 3. We are examining loci of plasticity both in vitro and in vivo (as relates to Aim 1c). We have two manuscripts that were favorably reviewed, now being revised for resubmission, involving recordings in rat primary auditory cortex, frontal cortex, and locus coeruleus during the auditory behavior used in this application (Carcea et al., *Nature Communications* under revisions; Insanally et al., *Nature* under revisions).

Froemke RC, Carcea I, Barker AJ, Yuan K, Seybold B, Martins ARO, Zaika N, Bernstein H, Wachs M, Levis PA, Polley DB, Merzenich MM, Schreiner CE. Long-term modification of cortical synapses improves sensory perception. *Nature Neuroscience* 2013; 16:79-88. PMC: 3711827

Carcea I, Froemke RC. Cortical plasticity, excitatory-inhibitory balance, and sensory perception. *Progress in Brain Research* 2013; 207, 65-90. PMID: 24309251 PMC: 4300113

Martins ARO, Froemke RC. Coordinated forms of noradrenergic plasticity in the locus coeruleus and primary auditory cortex. *Nature Neuroscience* 2015; 18:1483-1492. PMID:26301326 PMC: 4583810

Marlin BJ, Mitre M, D'amour JA, Chao MV, Froemke RC. Oxytocin enables maternal behaviour by balancing cortical inhibition. *Nature* 2015; 520:499-504. PMID: 25874674 PMC: 4409554

D'amour JA, Froemke RC. Inhibitory and excitatory spike-timing-dependent plasticity in the auditory cortex. *Neuron* 2015; 86; 514-528. PMID: 25843405 PMC: 4409545

Ponte Costa R, Froemke RC, Sjöström PJ, van Rossum MC. Unified pre- and postsynaptic long-term plasticity enables reliable and flexible learning. *eLife* 2015; 4. PMID: 26308579 PMC: 4584257

Froemke RC. Plasticity of cortical excitatory-inhibitory balance. *Annual Review of Neuroscience* 2015; 38, 195-219. PMID: 25897875 PMC: 4652600

Froemke RC, Schreiner CE. Synaptic plasticity as a cortical coding scheme. *Current Opinion in Neurobiology* 2015; 35, 185-199. PMID: 26497430 PMC: 4641776

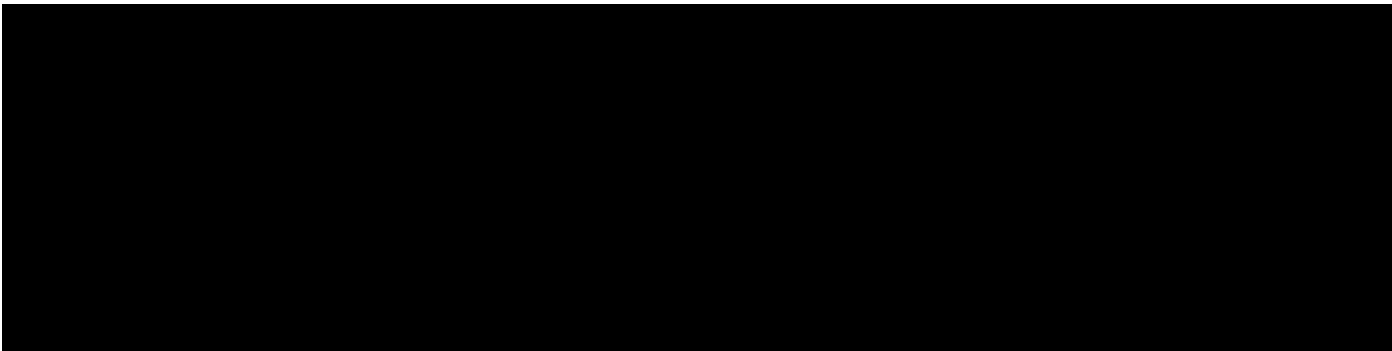
King J, Insanally M, Jin M, Martins ARO, D'amour JA, Froemke RC. Rodent auditory perception: critical band limitations and plasticity. *Neuroscience* 2015; 296, 55-65. PMID:25827498 PMC: 4426073

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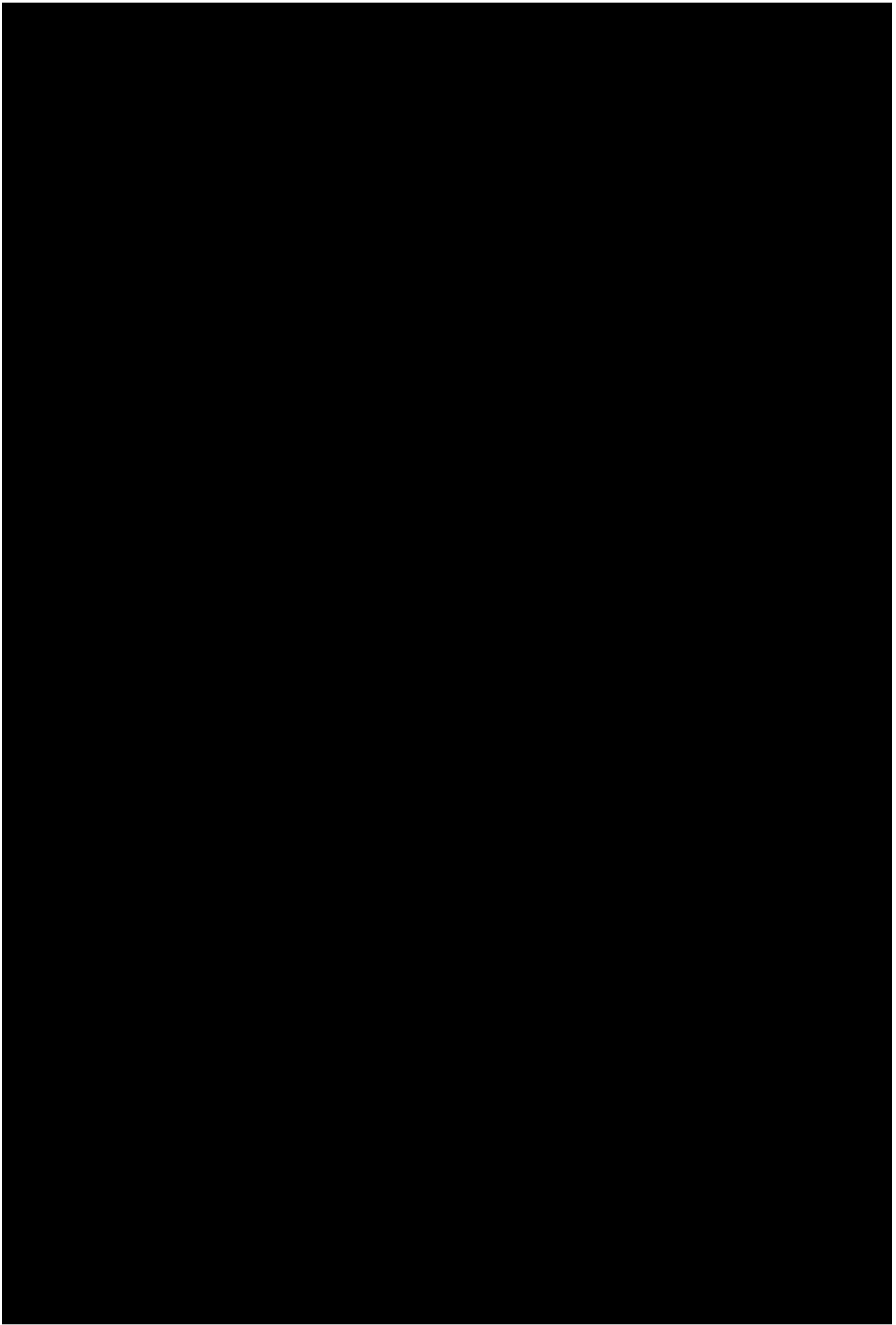
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Authentication of Key Resources Plan

All resources, chemicals, and reagents used for the experiments in this proposal are standard and supported by numerous publications (e.g., APV to block NMDA receptors, pharmacological reagents, the ChETA variant of channelrhodopsin-2, tdTomato interneuron transgenic mouse lines).