Award Number: W81XWH-19-1-0251

TITLE: Systems genetics of Tuberous Sclerosis Complex outcomes using BXD recombinant inbred mice

PRINCIPAL INVESTIGATOR: John Matthew Mahoney

CONTRACTING ORGANIZATION: University of Vermont, Larner College of Medicine

REPORT DATE: July 2020

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

DISTRIBUTION STATEMENT: Approved for Public Release;
Distribution Unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.

REPORT DOCUMENTATION PAGE			*Form Approved* *OMB No. 0704-0188*

Public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing this collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden to Department of Defense, Washington Headquarters Services, Directorate for Information Operations and Reports (0704-0188), 1215 Jefferson Davis Highway, Suite 1204, Arlington, VA 22202-4302. Respondents should be aware that notwithstanding any other provision of law, no person shall be subject to any penalty for failing to comply with a collection of information if it does not display a currently valid OMB control number. **PLEASE DO NOT RETURN YOUR FORM TO THE ABOVE ADDRESS.**

1. REPORT DATE	**2. REPORT TYPE**	**3. DATES COVERED**
JULY 2020	Annual	1 July 2019 – 30 June 2020

4. TITLE AND SUBTITLE

Systems Genetics of Tuberous Sclerosis Complex Outcomes Using BXD Recombinant Inbred Mice

5a. CONTRACT NUMBER W81XWH-19-1-0251

5b. GRANT NUMBER TS180087

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)

John Matthew Mahoney

E-Mail: john.m.mahoney@uvm.edu

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

University of Vermont Larner College of Medicine
89 Beaumont Ave.
Burlington, VT 05405-0068

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)

U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT

Approved for Public Release; Distribution Unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

The goal of this hypothesis development project is to demonstrate that diversity in genetic background significantly influences tuberous sclerosis complex (TSC) outcomes in mice. This will show that it is possible to genetically map susceptibility and resilience genes to different classes of TSC outcomes, including TSC-associated neurological disorders, epilepsy, and brain malformations. During this funding period we learned that the available *Tsc1*-mutant mouse model has a mixed genetic background, meaning that its genome is a combination of multiple inbred strain genomes. This introduces unwanted genetic variability into the breeding scheme for our experiments. To overcome this, we have undertaken a backcross breeding program to purify the genetic background of the *Tsc1*-mutant mouse to eliminate this variability in future experiments. The significant development to date is that we have finished breeding the second generation of this backcross, resulting in a mouse with an approximately 75% pure genome. We will use these mice to make *Tsc1* knockouts and go forward with those knockouts to study the effects of genetic background on TSC outcomes. We will complete the backcross breeding scheme to create a mouse with around 95% purity in parallel with other experiments.

15. SUBJECT TERMS

Systems genetics; TSC-associated neuropsychiatric disorders; epilepsy; histopathology; mouse model

16. SECURITY CLASSIFICATION OF:			**17. LIMITATION OF ABSTRACT**	**18. NUMBER OF PAGES**	**19a. NAME OF RESPONSIBLE PERSON USAMRDC**
a. REPORT	**b. ABSTRACT**	**c. THIS PAGE**			
U	U	U	UU	14	USAM TELEPHONE NUMBER

Table of Contents

	Page
INTRODUCTION………………………………………………………….………..	4
KEYWORDS………………………………………………………………………..	4
ACCOMPLISHMENTS…………………………………….……………………	4-6
IMPACT…………………………………………………………………………....	6
CHANGES/PROBLEMS………………………………………………………..	6-8
PRODUCTS………………………………………………………………………	8-9
PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS …………	9-13
REFERENCES…………………………………………………………………...	14

1. INTRODUCTION

The **subject** of this project is the development of novel mouse models for tuberous sclerosis complex (TSC), called BXD-TSC mice. The **purposes** of this project are to demonstrate that different genetic backgrounds exhibit heritable variation in TSC-associated outcomes and to establish a novel panel of mouse models capturing this diversity. Briefly, we are leveraging the existing BXD mouse diversity panel, which is a well-established collection of inbred mouse strains with high neurophenotypic diversity, to generate mice that carry one copy of a *Tsc1* loss-of-function allele inherited from their mother with a BXD genotype inherited from their father. Because the BXD genomes are combinations of the B6 and DBA/2J (D2) genomes, and are fully sequenced, this panel will enable genetic mapping of sensitivity and resilience alleles for behavioral, electrohysiological, and histological outcomes in TSC. The **scope** of this project is to develop the hypothesis that outcome heterogeneity exists among the BXDs by testing a subset of six selected BXD strains (out of ~200 available strains) along with the parent B6 and D2 strains. In these eight strains (six BXD + B6 + D2), our aims are to measure behavioral traits associating with anxiety, sociability, learning and memory, followed by electrophysiological screening for seizures and other epileptiform activity, and finally measure histopathological features of brain anatomy. With these measurements, we will be able to estimate the heritability of these traits as a prelude to a full genetic mapping experiment on a comprehensive panel with high genetic resolution.

2. KEYWORDS:

Systems genetics; TSC-associated neuropsychiatric disorders; epilepsy; histopathology; mouse model

3. ACCOMPLISHMENTS:

a. What were the major goals of the project?

The major goals outlined in the SOW were the following specific aims, each with associated Major Tasks (below):

Specific Aim 1: Genetically map differences in TSC-associated behaviors of BXD-TSC mice.

Specific Aim 2: Genetically map differences in the seizure outcomes of BXD-TSC mice.

Specific Aim 3: Genetically map differences in the histopathological outcomes of BXD-TSC mice.

b. What was accomplished under these goals?

Specific Aim 1

Major Task 1: Regulatory approval (Target: By month 6; Progress: 100%)

We received final regulatory approval from ACURO to begin this project on December 10, 2019.

Major Task 2: Establish breeding colony (Target: By month 8; Progress: 75%)

Since January 28, 2020, when money was released to our lab by UVM to begin experiments, the major development has been that we learned that the commercially available conditional *Tsc1* knock out (cKO) mouse strain, called the *Tsc1*^{Tm1Djk} strain (https://www.jax.org/strain/005680), does not have a pure, inbred background. In particular, due to its derivation as a transgenic mouse, its genome is a combination of the 129S4/SvJae (129) strain and the B6 strain. This has forced us modify our approach to the specific aims by backcrossing the *Tsc1*^{Tm1Djk} mouse to the B6 strain (described in detail in section 5a below). We have currently bred the N2 generation of the backcross, which has ~75% B6 genome, and we are now using these mice to create the heterozygous *Tsc1* KO mice (N2-*Tsc1*$^{+/-}$ mice), which will be used for our breeding scheme in the SOW. To make N2-*Tsc1*$^{+/-}$ mice requires one more round of breeding with the CMV-Cre mouse (https://www.jax.org/strain/006054) to create germline *Tsc1*-KO mice. This puts us on track to finish setting up our breeding colony in ~8-10 weeks (accounting for breeding, gestation, and maturation).

In order to complete this work, I have hired a lab technician (Khalil Abedrabbo) at 25% effort. Because his current role only involves colony management for the backcross, I have not hired him at the budgeted 50% effort. I have also recruited a graduate student (Montana Lara) whose PhD thesis will be on this project. Ms. Lara is independently funded through her program and will not draw salary from this award, but she will complement Mr. Abedrabbo with husbandry and phenotyping throughput in the subsequent funding period.

Major Task 3: Behavioral phenotyping (Target: By month 20; Progress: 0%)
Our lab technician, Mr. Abedrabbo, has extensive experience with animal behavioral phenotyping, including extensive work with macaques at Yale and rodents at UT Dallas. Our graduate student, Ms. Lara, has been fully trained on our behavioral protocols. They will be able to complete Specific Aim 1 as described in the SOW as soon as BXD-TSC mice are ready.

Specific Aim 2
Major Task 1: Video-EEG monitoring (Target: By month 20; Progress: 0%)
When BXD-TSC mice are ready and have completed the behavioral pipeline (Aim 1), we will implant them with wireless EEG electrodes and monitor them for 5 days to screen for seizure activity and epileptiform discharges.

There have been two major developments on this aim. First, we have tested and optimized a new wireless EEG system from Pinnacle Systems to establish that we can detect epileptiform discharges and frank seizures on this system (Fig. 1). This wireless system allows freer movement of the mice during monitoring, has fewer hazards (e.g. shorting wires with drinking water), and fewer obstructions to video monitoring.

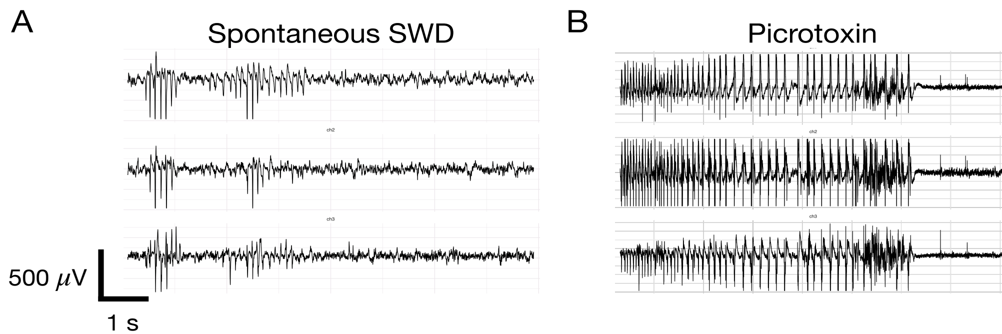

Figure 1. Epileptiform activity recorded in DBA/2J (D2) wild type mice. A) We recorded multiple spontaneous spike-and-wave discharges (SWDs) in D2 mice. An example SWD recapitulates the canonical 3 Hz form. B) Using picrotoxin, we observed frank seizures as an additional positive control.

The second development is that both Ms. Lara and Mr. Abedrabbo have been trained and have successfully implanted Pinnacle electrodes into mice and recorded biological signals. In particular, Ms. Lara has validated a report from the literature that D2 mice have frequent spike-and-wave discharges [Fig. 1A; *cf.* (Letts, Beyer, and Frankel 2014)]. Consistent with the literature, we have not seen any such events in B6 mice.

As a further positive control for frank seizures, we have also recorded a mouse that was injected with picrotoxin to induce a behavioral seizure (Fig. 1B). These results demonstrate that we can detect both behavioral and sub-behavioral seizures. Ms. Lara has since begun to write automated software to detect epileptiform discharges based on their spectral content. Over the next funding period, she will continue to develop this pipeline in parallel with husbandry and data collection. We are now fully staffed and trained to complete Specific Aim 2 as described in the SOW as soon as BXD-TSC mice are ready.

Specific Aim 3
Major Task 1: Histological analysis (Target: By month 20; Progress: 0%)
When BXD-TSC mice are ready and have completed the behavioral and electrophysiological pipelines (Aims 1 & 2), we will sacrifice them and prepare histological sections.

Mr. Abedrabbo and Ms. Lara have both received appropriate training on preparing histological sections and are ready to complete this aim as outlined in the SOW. We are now fully staffed and trained to complete Specific Aim 3 as described in the SOW as soon as BXD-TSC mice are ready.

c. What opportunities for training and professional development has the project provided?

Nothing to Report

d. How were the results disseminated to communities of interest?

Nothing to report.

e. What do you plan to do during the next reporting period to accomplish the goals?

As described above, over the next few months, we will be in a position to use N2-*Tsc1*$^{+/-}$ mice to breed to B6, D2, and BXD backgrounds to generate BXD-TSC mice. We are also fully staffed and trained to perform all relevant phenotyping. Over the next funding period, we will run our phenotyping pipeline in *waves* of two strains at a time. We will begin with the B6 and D2 strains. As soon as B6 and D2 litters are weaned, we will breed the N2-*Tsc1*$^{+/-}$ mice to two BXD strains, and repeat until all eight strains are complete. Note that, if our hypotheses are correct, the B6 and D2 strains will significantly differ in at least some TSC outcomes, demonstrating that TSC outcomes are genetically mappable in the BXD panel. This outcome is publishable and is sufficient preliminary data to support a larger grant application to perform a large-scale genetic mapping experiment (the desired outcome of this hypothesis development grant). Upon completing the B6/D2 wave, we will immediately prepare a manuscript for a journal and an extramural grant proposal.

4. IMPACT:

a. What was the impact on the development of the principal discipline(s) of the project?

Nothing to Report

b. What was the impact on other disciplines?

Nothing to Report

c. What was the impact on technology transfer?

Nothing to Report

d. What was the impact on society beyond science and technology?

Nothing to Report

5. CHANGES/PROBLEMS:

a. Changes in approach and reasons for change

As discussed above, we learned that the commercially available *Tsc1* conditional KO mouse, *Tsc1*^{Tm1Djk}, has an unknown mixed background that is not purely B6. This caused us to modify our approach. Because the goal of this project is to develop the hypothesis that TSC outcomes can be genetically mapped in the BXD panel, we also ultimately require a *Tsc1* KO mouse on a pure B6 background to ensure an inbred mouse after Cre-mediated KO of *Tsc1*. Thus, we decided to begin a backcross breeding program to put the floxxed *Tsc1* allele of the *Tsc1*^{Tm1Djk} strain onto a pure B6 background [*cf.* (Silver 1995)].

Briefly, in a backcross you mate the transgenic animal (*Tsc1*^{Tm1Djk} in this case) to the target background (B6 in this case). These progeny are called the N1 generation and they are crossed back to the pure B6 strain to make

the N2 generation. Half of the N2 mice inherit the floxxed *Tsc1* allele, which we ascertain with PCR, and these are crossed back to B6 strain to make generation N3, and so on. The amount of B6 genome increases monotonically with each generation. To speed the process, we are using marker-based selection, where we genotype each generation to select individuals with the most B6 in their genome to continue the backcross. We assay the amount of B6 genotype using a SNP genotyping array at The Jackson Labroatory Genome Scanning Service (https://www.jax.org/jax-mice-and-services/breeding-and-rederivation-services/genome-scanning). At the culmination of this process (N4 or N5), we will have a mouse with >93% B6 background, and a genetic map of where residual 129 strain sequence still resides. As soon as this mouse is available, we will donate it to The Jackson Laboratory for dissemination to the worldwide research community (https://www.jax.org/jax-mice-and-services/cryo-and-strain-donation/donate-a-strain). This mouse will be of widespread interest to the TSC community, because it will enable genetically controlled crosses with Cre lines on a pure B6 background. Such mice are infinitely reproducible genetically, so there is no confound due to recombination. Critically, this mouse will also support our follow up grant applications as a novel and powerful reagent for a large-scale genetic mapping study of TSC outcomes.

The major drawback to the backcross is that it is time consuming, even using a marker-based selection strategy. In order to complete our study objectives in a timely manner, we have decided to undertake the SOW aims using N2-*Tsc1*$^{+/-}$mice instead of waiting for the fully purified B6-*Tsc1*$^{+/-}$ mouse that we will have at the end of the backcross program. While this will include some uncontrolled genetic variability into our assays due to recombination mixed background N2-*Tsc1*$^{+/-}$ strain and the B6 background of the CMV-Cre mouse, we expect this variance to be small compared to the strain effects in the BXD-TSC lines. Indeed, we expect the *Tsc1* KO and strain-background effects to dominate the effects of residual 129 genome. We will also control for this effect statistically when we perform analyses. Based on prior literature and our power calculations, we expect to retain statistical power with our current planned numbers to identify genotype-by-strain interaction as describe in the original application.

We have just begun breeding the N2 mice to the CMV-Cre mice and expect N2-*Tsc1*$^{+/-}$ to be ready for breeding B6 and D2 mice for the first wave of experiments in the next 2-3 months. As we progress through the backcross, and depending on the amount B6 genome, we will consider creating N3-*Tsc1*$^{+/-}$ mice, etc., that we will use with later waves of BXD strains.

In the end, this modified approach will both establish the hypotheses of this project and deliver a novel conditional knockout mouse on a standard inbred background. We expect the backcross to be complete by December 2021, within two years since the start of spending authorization in January 2020.

b. Actual or anticipated problems or delays and actions or plans to resolve them

There have been three delays:

First, after regulatory approval in December, there was a one-month delay before UVM released a chart string for our group to begin spending on this award. We received a chart string on January 28, 2020.

The second delay has been the overhead due to researching the genetic background of the *Tsc1*^{Tm1Djk} mouse, planning and then beginning to execute the backcross (see section 5a).

Third, the COVID-19 lockdown dramatically lowered our in-lab time from March to June. However, we were able to maintain a breeding schedule and have now completed the N2 generation and are beginning alternative approach described above. From this point forward, the backcross and the SOW work can be performed in parallel.

In total, we estimate that we are ~7 months behind schedule on establishing our breeding colony as described in the SOW.

c. Changes that had a significant impact on expenditures

There have been two significant impacts on expenditures:

First, because of preparing for the backcross, there was a delay in purchasing any mice or animal care for this project.

Second, we did not hire Mr. Abedrabbo on this project until June 1, 2020, because Ms. Lara was still preparing the backcross scheme and then the COVID-19 lockdowns. We have also opted to have his effort at 25%, rather than the budgeted 50%. This is because Ms. Lara will be able to perform most of the experiments. Because Ms. Lara is funded through her graduate program over the next funding period, her effort relieves our budget for tasks that Mr. Abedrabbo would otherwise undertake. Mr. Abedrabbo's responsibilities are now limited to animal husbandry and aiding Ms. Lara with experimental preparations. We anticipate that the reduction in spending on Mr. Abedrabbo's effort will compensate the higher costs for animal care for the backcross.

d. Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents

Nothing to Report

e. Significant changes in use or care of human subjects

Nothing to Report

f. Significant changes in use or care of vertebrate animals.

Nothing to Report

g. Significant changes in use of biohazards and/or select agents

Nothing to Report

6. PRODUCTS:

a. Publications, conference papers, and presentations

i. **Journal publications.**

Nothing to Report

ii. **Books or other non-periodical, one-time publications.**

Nothing to Report

iii. **Other publications, conference papers, and presentations.**

Nothing to Report

b. Website(s) or other Internet site(s)

Nothing to Report

c. **Technologies or techniques**

Nothing to Report

d. **Inventions, patent applications, and/or licenses**

Nothing to Report

e. **Other Products**

Nothing to Report

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

a. **What individuals have worked on the project?**

Name:	J. Matthew Mahoney
Project Role:	PI
Researcher Identifier (e.g. ORCID ID):	0000-0003-1425-5939
Nearest person month worked:	1
Contribution to Project:	Dr. Mahoney hired staff, trained staff and mentees, and directed experiments.
Funding Support:	DOD TS180087 NIH R21AI145306 P20GM130454-01 R21NS117112 R21LM012615

Name:	Montana Lara
Project Role:	Graduate Student
Researcher Identifier (e.g. ORCID ID):	0000-0001-5810-4425
Nearest person month worked:	6
Contribution to Project:	Ms. Lara has designed our backcross breeding scheme, optimized our wireless electrophysiology system, and trained on behavior and histopathology.
Funding Support:	NIH R21AI145306

Name:	Rodney C. Scott
Project Role:	Co-I
Researcher Identifier (e.g. ORCID ID):	0000-0002-8912-7475
Nearest person month worked:	1
Contribution to Project:	Dr. Scott helped design and oversee experiments and training.
Funding Support:	DOD TS180087 Great Ormond Street Charity 178391 Mallinckrodt Enterprises, LLC Services Agreement NIH R21NS117112 R01NS108765 R01NS110945

b. Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?

PI Mahoney and Co-I Scott have had multiple changes to active support (described below). These changes have not affected effort on this project.

MAHONEY, J.M.
New active support:
R21NS117112 (Scott) 4/1/20-3/31/22 0.6 CAL MOS
NIH/NINDS $150,000
Brain stimulation for prevention of epileptogenesis
The overall goal of this project is to establish the time course for developing abnormalities in hippocampal neural circuit action potential firing dynamics after pilocarpine induced hippocampal injury, determine whether maintenance of normal neuronal firing dynamics prevents epileptogenesis and establish the genetic mechanisms of those effects.
Role: Co-I

R21 AI145306 (Krementsov) 07/01/19-06/30/21 0.6 CAL MOS
NIH/NIAID $275,000 (2 years; total direct costs)
Next generation systems analysis of pathogenetic mechanisms underlying CNS autoimmunity
The goal of this project is to utilize the Collaborative Cross mouse model to identify novel genes controlling various aspects of disease pathogenesis in a mouse model of multiple sclerosis (MS) (Aim 1), and to use bioinformatic approaches to identify novel candidate genes, epistatic interactions, and human homologous genes relevant to MS (Aim 2).
Role: Co-Investigator

Note: co-funding with RG-1807-31533, below.

RG-1807-31533 (Krementsov) 10/01/19-09/31/22 0.0 CAL MOS

NMSS $284,434 (3 years; total direct costs)

Next generation systems analysis of pathogenetic mechanisms underlying CNS autoimmunity using the Collaborative Cross

The goal of this project is to utilize the Collaborative Cross (CC) mouse model to identify novel genes controlling various aspects of disease pathogenesis in a mouse model of multiple sclerosis (MS) (Aim 1). Additionally, in Aim 2, we will determine the immune-pathologic mechanisms underlying any newly identified genetically controlled phenotypes in CC mice, and as well as mechanisms underlying genetically-controlled sex differences in disease pathogenesis.

Role: Co-Investigator

Note: co-funding with R21 AI145306, above.

P20GM130454-01 (Whitfield) 12/1/19-11/30/23 6.0 CAL MOS

Dartmouth College (sub) / NIH (primary)

Center for quantitative biology: a focus on “omics” from organisms to single cells

The long-term goal of this proposal is to develop computational methods to identify tissue-specific molecular pathways underlying complex disease. Dr. Mahoney’s research project proposes to develop and test a computational framework called functional genomic denoising (FGD) that uses transciptomic data to computationally predict tissue-specific disease pathways in settings where major genes are not differentially expressed and where relevant tissues cannot be assayed.

Role: Project PI, Subaward PI

Changes to other support:
R21LM012615 (Tyler/Mahoney) 6/5/18-5/31/21 3.69 CAL MOS
NIH/NLM NCE
Inferring molecular mechanisms of complex disease by integrating patterns of epistasis with functional genomic networks
The overall goal of this proposal is to develop computational methods that will identify genes driving epistasis between quantitative trait loci.
Role: PI (MPI)
R21LM012615 entered its No Cost Extension period effective 6/1/20. Its end-date has been extended accordingly.

R21NS117112 (Scott, PI; Mahoney, Co-I) was recently awarded with an effective date of 4/1/2020.

R21 AI145306 (Krementsov, PI; Mahoney, Co-I) recently had a COVID-19 budget reduction. Dr. Mahoney has dropped his effort to 0% starting 07/01/2020.

SCOTT, R.C.

New active support:

R21NS117112 (Scott) 4/1/20-3/31/22 2.4 CAL MOS
NIH/NINDS $150,000
Brain stimulation for prevention of epileptogenesis
The overall goal of this project is to establish the time course for developing abnormalities in hippocampal neural circuit action potential firing dynamics after pilocarpine induced hippocampal injury, determine whether maintenance of normal neuronal firing dynamics prevents epileptogenesis and establish the genetic mechanisms of those effects.
Role: PI

Services Agreement (Scott) 2/1/2020-1/31/2021 1.2 CAL MOS
Mallinckrodt Enterprises, LLC $163,540
Systems level mechanisms of ACTH efficacy
This project aims to (1) characterize acute changes in gene expression networks following 60 early life seizures in rats; and (2) examine long term effects of Acthar gel on gene networks and in-vivo action potential firing dynamics.
Role: PI

R01NS108765 (Holmes) 7/15/18-3/31/23 1.8 CAL MOS
NIH/NINDS $243,375
Mechanisms of cognitive impairment following early-life seizures
The goals of this project are to (1) assess temporal coding of action potentials (APs) in the intra-HPC (CA3-CA1) and CA1-mPFC networks following ELS; (2) determine if remedial training in rats following ELS improves cognition as well as coding abnormalities within and between HPC and mPFC networks; and (3) determine if optogenetic stimulation of the MS can alter temporal coding in HPC and mPFC.
Role: Co-Investigator

R01NS110945 (Weston) 2/1/19-1/31/24 0.6 CAL MOS
NIH/NINDS $222,089
Synaptic changes in hypersynchronous network activity in mTORopathies
The goals of this project are to (1) determine whether multiple mTORopathy models share common synaptic alterations; (2) assess the contributions of synaptic and morphological changes to hypersynchronous activity and epilepsy in mTORopathy models; and (3) delineate the spatiotemporal relationship between synaptic changes and hypersynchronous network activity in mTORopathy models.
Role: Co-Investigator

178391 (Scott) 1/1/2019-12/21/2021 1.2 CAL MOS
Great Ormond Street Hospital Children's Charity £300,000.00
Novel network analysis of intracranial stereoelectroencephalography
The goal is to characterize interictal abnormalities in single unit neural dynamics and to establish whether the regions that display abnormal dynamics are consistent with the epileptogenic zone. This study will be carried out in children undergoing invasive recordings as part of work up for epilepsy surgery.
Role: PI

Changes to other support:

R21NS117112 (Scott, PI) was recently awarded with an effective date of 4/1/2020.

c. **What other organizations were involved as partners?**
Nothing to Report

REFERENCES

Letts, V. A., B. J. Beyer, and W. N. Frankel. 2014. “Hidden in Plain Sight – Spike-Wave Discharges in Mouse Inbred Strains.” *Genes, Brain, and Behavior* 13 (6): 519–26. https://doi.org/10.1111/gbb.12142.

Silver, L. M. 1995. “Mouse Genetics: Concepts and Applications.” *Mouse Genetics: Concepts and Applications.* https://www.cabdirect.org/cabdirect/abstract/19960104116.