AWARD NUMBER: W81XWH-21-1-0870

TITLE: Acute to Chronic Pain Signatures in Traumatic Injury

PRINCIPAL INVESTIGATOR: Jennifer E. Nyland, PhD

CONTRACTING ORGANIZATION: Pennsylvania State University, Milton S. Hershey Medical Center, Hershey, PA

REPORT DATE: September 2023

TYPE OF REPORT: Annual

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

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE	2. REPORT TYPE	3. DATES COVERED
September 2023	Annual	01Sep 2022 - 31 Aug 2023

4. TITLE AND SUBTITLE

Acute to Chronic Pain Signatures in Traumatic Injury

5a. CONTRACT NUMBER

5b. GRANT NUMBER
W81XWH-21-1-0870

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)
Jennifer E. Nyland

E-Mail: jnyland@pennstatehealth.psu.edu

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

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

Pennsylvania State University, The
DBA Controller's Office for the College of Medicine
500 University AVE RM C1607
Hershey, Pennsylvania 17033-2360

8. PERFORMING ORGANIZATION REPORT

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

U.S. Army Medical Research and Development 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

Persistent and chronic pain are significant contributors to diminished quality of life following acute musculoskeletal injury. The goal of this project is to identify critical biomarkers to predict susceptibility/resilience to the development of chronic pain after a traumatic injury. Here, we are using a population of patients with blunt chest trauma with multiple (>2) closed rib fractures, as our published prospective data indicate the chronification of pain after multiple rib fractures observed in our institution ranges from 40 to 50%. Biomarkers assessed include 1) prognostic clinical factors that predict a predisposition for susceptibility and/or resilience to pain chronification; 2) functional brain imaging biomarkers associated with the development of chronic pain; and 3) nociceptive genetic patterns at the time of injury that are predictive of pain chronification.

15. SUBJECT TERMS

Pain; Trauma; Biomarkers

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			USAMRDC
Unclassified	Unclassified	Unclassified	Unclassified	16	19b. TELEPHONE NUMBER *(include area code)*

TABLE OF CONTENTS

1. INTRODUCTION:

Persistent and chronic pain are significant contributors to diminished quality of life following acute musculoskeletal injury. The goal of this project is to investigate chronification of acute pain in patients with blunt chest trauma with multiple (>2) closed rib fractures by assessing psychological, biological, and functional neural connectivity biomarkers during the transition from acute to chronic pain. Rib fractures are commonly recognized as a significant source of acute pain following injury; however, they also have a high rate of transition to chronic pain. Our published prospective data indicate the chronification of pain after multiple rib fractures observed in our institution ranges from 40 to 50%. There remains a large gap in understanding of how acute pain transitions to chronic pain and how clinical measures can predict pain chronification. The objective of this study is to identify critical biomarkers to predict susceptibility/resilience to the development of chronic pain using blunt chest trauma with multiple rib fractures as a model. As such, we intend to 1) identify prognostic clinical factors that predict a predisposition for susceptibility and/or resilience to pain chronification; 2) identify functional brain imaging biomarkers associated with the development of chronic pain; and 3) identify nociceptive genetic patterns at the time of injury that are predictive of pain chronification.

2. KEYWORDS:

Chronic Pain; Acute Pain; Trauma; Chronification

3. ACCOMPLISHMENTS:

What were the major goals of the project?

Statement of Work (items in **blue** are either complete or currently ongoing)

Task	Time	Proposed/Actual Dates	
Major Task 1: Study Startup and Regulatory Approval	**Months**	**Proposed**	**Actual**
Subtask 1: Compose and submit protocol for IRB approval	1	9/30/2021	8/4/2021
Subtask 2: Submit and obtain HRPO approval	1	9/30/2021	9/28/2021
Subtask 3: Develop Manual of Operations for study team	2-3	11/30/2021	10/28/2021
Subtask 4: Develop REDCap database and data entry parameters	2-3	11/30/2021	10/20/2021
Milestone(s) Achieved: IRB/HRPO-approved protocol	3	11/30/2021	9/28/2021

Statement of Work Continued

Specific Aim 1: Identify prognostic clinical factors that predict a predisposition for susceptibility and/or resilience to pain chronification.

Task	Time	Proposed/Actual Dates	
Major Task 2: Identify clinical indicators that predict chronic pain in patients with multiple rib fractures	**Months**	**Proposed**	**Actual**
Subtask 1: Participant screening for eligibility	3-32	2/28/2023	Ongoing
Subtask 2: Informed consent and enrollment of ~150 pts	3-32	2/28/2023	34% Complete
Subtask 3: Extraction of patient records from EMR	3-32	2/28/2023	34% Complete
Subtask 4: Pain assessments and QST	3-32	2/28/2023	34% Complete
Subtask 5: Submit manuscript on clinical indicators of chronic pain	36	8/31/2024	NA
Milestone(s) Achieved: Baseline pain characteristics, determination of clinical predictors of chronic pain	36	6/30/2024	NA

Task	Time	Proposed/Actual Dates	
Major Task 3: Complete follow-up assessments and identify psychological factors that predict chronic pain	**Months**	**Proposed**	**Actual**
Subtask 1: 3-month follow-up assessments	6-33	5/31/2024	28% Complete
Subtask 2: 6-month follow-up assessments	9-36	8/31/2024	27% Complete
Subtask 3: Verification of chronic pain	9-36	8/31/2024	27% Complete
Subtask 4: Analysis/interpretation of psychological assessments	7-36	8/31/2024	27% Complete
Subtask 5: Submit manuscript on psych indicators of chronic pain	36	8/31/2024	NA
Milestone(s) Achieved: Determination of chronic pain outcomes, psychological predictors of chronic pain	36	6/30/2024	NA

Specific Aim 2: Identify functional brain imaging biomarkers associated with development of chronic pain.

Task	Time	Proposed/Actual Dates	
Major Task 4: Identify differences in functional connectivity that predict chronic pain	**Months**	**Proposed**	**Actual**
Subtask 1: Development of fMRI algorithms and protocol	1	9/30/3021	10/12/2021
Subtask 2: Completing fMRI scans at enrollment	3-32	2/28/2024	32% Complete
Subtask 3: Cleaning of images	4-34	4/30/2024	29% Complete
Subtask 4: Analysis and interpretation of data	18-36	8/31/2024	NA
Subtask 5: Statistical analysis of patient outcomes (from Aim 1) compared between chronic pain and non-chronic pain participants	22-36	8/31/2024	NA
Subtask 6: Submit manuscript on fMRI predictors of chronic pain	36	8/31/2024	NA
Milestone(s) Achieved: fMRI assessments for all patients and the time of injury, association of connectivity with pain outcomes, functional biomarkers of pain chronification	36	8/31/2024	NA

Specific Aim 3: Identify nociceptive genetic pattern at the time of injury that are predictive of pain chronification.

Task	Time	Proposed/Actual Dates	
Major Task 5: (For each major task, define the key hypothesis or the main study(s) to be tested.)	**Months**	**Proposed**	**Actual**
Subtask 1: Blood collection at enrollment	3-32	2/28/2024	34% Complete
Subtask 2: Sample processing and storage	3-36	6/30/2024	34% Complete
Subtask 3: Sample analysis	16, 36	12/31/2022, 8/31/2024	24% Complete NA
Subtask 4: Statistical analysis of patient outcomes (from Aim 1) compared between chronic pain and non-chronic pain participants	22-36	8/31/2024	NA
Subtask 5: Submit comprehensive manuscript on combined findings	36	8/31/2024	NA
Milestone(s) Achieved: Genomic analysis for all patients that completed all 3 assessments and the time of injury	36	8/31/2024	NA

What was accomplished under these goals?

1) Major Activities: Major activities during this reporting period include continued enrollment and follow-up of study participants. We have also focused on the initial cleaning and processing of fMRI images for all participants. Further, preliminary data analyses have been conducted to confirm that the study will be appropriately powered to detect differences between participants who develop chronic pain and those who do not. Genetic analyses have been conducted for the first 20 participants. We have also completed the extraction of health history from the EMR and Trauma Registry for the first 32 participants. Importantly, we have not lost a single participant to follow-up. All follow-up assessments for 3- and 6-month follow-ups have been completed and verified in our REDCap database.

2) Specific Objectives: During this period, the objectives were focused on participant enrollment and continued follow-up.

3) Significant Results or Key Outcomes: The fMRI data have undergone preliminary analyses to determine if our proposed recruitment goal will yield sufficient power to determine differences between groups. An initial analysis of 20 participants yielded significant correlations in connectivity of the default mode network and scores on the McGill Pain Questionnaire. This gives us confidence that we will be powered to conduct our analyses with a lower number of participants than originally proposed. We intend to conduct an interim analysis in the coming months to determine if our target enrollment can be revised.

4) Other Achievements: Based on the fMRI and initial study visit assessments, we submitted and presented two abstracts for scientific conferences; one for the MHSRS conference and one for the International Society for Magnetic Resonance in Medicine annual meeting.

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

Nothing to report.

How were the results disseminated to communities of interest?

Nothing to report.

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

Our goals and objectives over the next reporting period include ramping up recruitment activities and continuing data collection and subject enrollment. To boost recruitment, we will continue to send invitation letters to potential participants that were discharged before a study team member could discuss the study with them. We also have modified the inclusion/exclusion criteria to allow ≥ 1 rib fractures (previously was ≥ 2 rib fractures). We believe this will expand our pool of potential participants without altering the study goal. We decided to make this change for two reasons. First, during recruitment, several potential participants were excluded because x-ray confirmation indicated only one fractured rib. These patients reported pain similar to patients with 2 or more rib fractures. Second, we have also recognized that potential participants with greater than 2 fractured ribs are more likely to have more severe injuries overall, including TBI and spinal trauma. These patients are excluded as a result of their injury severity. We believe expanding our inclusion criteria to include participants with one or more fractured ribs will significantly benefit our enrollment. We have additionally created an option for participants with claustrophobia or metal that would interfere with the fMRI scan to opt out of the MRI and QST testing. This will allow us to collect blood specimens and participant-reported data for more participants, as genetic analyses require higher sample sizes than do imaging samples.

4. IMPACT:

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

Nothing to report.

What was the impact on other disciplines?

Nothing to report.

What was the impact on technology transfer?

Nothing to report.

What was the impact on society beyond science and technology?

Nothing to report.

5. CHANGES/PROBLEMS:

Changes in approach and reasons for change

Because of low recruitment, and participants with severe injuries being unwilling to participate because of the severity of their injuries, we modified the inclusion/exclusion criteria to allow ≥ 1 rib fractures (previously was ≥ 2 rib fractures). We believe this will expand our pool of potential participants without altering the study goal. We decided to make this change for two reasons. First, during recruitment, several potential participants were excluded because x-ray confirmation indicated only one fractured rib. These patients reported pain similar to patients with 2 or more rib fractures. Second, we have also recognized that potential participants with greater than 2 fractured ribs are more likely to have more severe injuries overall, including TBI and spinal trauma. These patients are excluded as a result of their injury severity. We believe expanding our inclusion criteria to include participants with one or more fractured ribs will significantly benefit our enrollment. We have additionally created an option for participants with claustrophobia or metal that would interfere with the fMRI scan to opt out of the MRI and QST testing. This will allow us to collect blood specimens and participant-reported data for more participants, as genetic analyses require higher sample sizes than do imaging samples.

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

Slowed enrollment during winter months. Enrollment was slowed by the abnormally mild winter this year. We had very little snow/ice during the winter months, which we believe reduced the number of patients in the trauma ICU within our inclusion criteria. Anecdotally, we have seen fewer falls in younger patients and overall motor vehicle accidents than we typically see during winter. However, we have seen numerous falls among patients outside our age criteria.

Team member turnover. We had one team member, Dr. Josh Hazelton, move to a different institution. Dr. Hazelton was the trauma surgeon on the team who would introduce the study to potential participants. If interested, Dr. Hazelton would notify our study team to visit the potential participant for recruitment. Dr. Hazelton was replaced in the study by Dr. Ryan Staszak, also a trauma surgeon, but it took a few months for Dr. Staszak to be brought up to speed with recruitment. Now that Dr. Staszak has been familiarized with the study, we are confident that recruitment will be smoother, as it was when Dr. Hazelton was involved.

Anticipated: In the next reporting period, our main focus will continue to be recruitment and enrollment. There are currently no new anticipated problems, however, as mentioned in previous reports, the COVID-19 pandemic is not yet behind us, which means that additional closures and adjustments may be required to keep participants and study team members safe.

Changes that had a significant impact on expenditures

Nothing to report.

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

Significant changes in use or care of human subjects

PROTOCOL (1 of 1 total):

Protocol [HRPO Assigned Number]: E024482.1a

Title: Acute to Chronic Pain Signatures in Traumatic Injury

Target required for clinical significance: 120

Target approved for clinical significance: 150

SUBMITTED TO AND APPROVED BY:

- No new protocol documents were submitted during this reporting period. 9/22-8/23

STATUS:

(i) Number of subjects recruited/original planned target: 41/120
Number of subjects screened/original planned target: 314/no target for screening
Number of patients enrolled/original planned target: 40/120
Number of patients completed/original planned target: Initial visit 40/120; 3-Month Follow-up 34/120; 6-Month Follow-Up 32/120

(ii) Report amendments submitted to the IRB and USAMRMC HRPO for review:

- Modification #8 was submitted to the Penn State IRB on 9/13/2022 and approved on 10/3/2022. The protocol and consent form were modified slightly to allow injury-related health records to be obtained from the Penn State Trauma Registry in addition to the EMR. In addition, a statement was added indicating that if a blood specimen cannot be collected from a participant, two saliva collection kits will be used as a backup. These changes were approved on an expedited basis and determined to be non-significant changes that did not increase the risk to the participants nor change the direction of the project.
- Modification #9 was submitted and approved by the Penn State IRB on 9/14/2022. The purpose of this modification was to remove Dr. Joshua Hazelton from the study team as he was leaving the institution.
- Modification #10 was submitted and approved by the Penn State IRB 10/5/2022. The purpose of this modification was to add new team members Ann Sipe (Research Technician III) and Erin King (rotating graduate student).

- Modification #11 was submitted and approved by the Penn State IRB 10/25/2022. The purpose of this modification was to add a new team member, Dr. Ryan Staszak, as a replacement for Dr. Joshua Hazelton (removed with mod #9). Dr. Staszak is a trauma surgeon, and his role is to assist in participant recruitment.
- Modification #12 was submitted and approved by the Penn State IRB 10/25/2022. The purpose of this modification was to update the Patient Invitation Letter to include recent changes to study team members (Josh Hazelton/Ryan Staszak). In addition, the protocol was adjusted to state that we will collect blood using 10ml tubes, as supplies issues were limiting our ability to obtain the 7ml tubes needed for collection. These changes were approved on an expedited basis and determined to be non-significant changes that did not increase the risk to the participants nor change the direction of the project.
- Modification #13 was submitted to the Penn State IRB on 3/22/2023 and approved by the Penn State IRB 4/10/2023. The purpose of this modification was to update the inclusion criteria from 2 or more rib fractures to 1 or more rib fractures. This required changes to the following study documents: Patient Invitation Letter, Informed Consent Form, Protocol, and the Phone Script. These changes were approved by a full IRB board and then submitted to the USAMRDC Office of Human and Animal Research Oversight, Office of Human Research Oversight (OHRO) on 8/2/2023 along with documents from Modification #15. On 8/17/2023 OHRO responded that the "changes described in the amendment applications, MOD 13 and MOD 15, do not meet OHRO's reporting requirements and do not require OHRO review and approval prior to implementation. The documents will be added to the study file and no additional action is needed."
- Modification #14 was submitted to the Penn State IRB on 4/14/2023 and approved on 4/17/2023. This modification included minor corrections to the text to maintain congruence and clarity with the previous modification. These changes were approved on an expedited basis and determined to be non-significant changes that did not increase the risk to the participants nor change the direction of the project.
- Modification #15 was submitted to the Penn State IRB on 5/19/2023 and approved on 6/28/2023. This modification allows a second option to participate in the study that only includes the blood/saliva collection and survey assessments. The rationale behind creating a second option was that many potential participants do not wish to participate in the fMRI/sensory testing due to claustrophobia or embedded metal. Therefore, potential participants that are not interested in the full study that includes fMRI and quantitative sensory testing may be offered the less intensive procedures alone. Because more data is needed for genetic analyses than would be necessary for fMRI analyses, this ensures we capture as many participants as possible to meet our study objectives. This required that a second informed consent form be created (option 2). These changes were approved by a full IRB board and then submitted to the USAMRDC Office of Human and Animal Research Oversight, Office of Human Research Oversight (OHRO) on 8/2/2023 along with documents from Modification #13. On 8/17/2023 OHRO responded that the "changes described in the amendment applications, MOD 13 and MOD 15, do not meet OHRO's reporting requirements and do not require OHRO review and approval prior to implementation. The documents will be added to the study file and no additional action is needed."

(iii) Adverse event/unanticipated problems involving risks to subjects or others and actions or plans for mitigation:

There were no adverse events/unanticipated problems involving risks to subjects or others during this reporting period.

Significant changes in use or care of vertebrate animals

No animal use research has been proposed to complete the Statement of Work.

Significant changes in use of biohazards and/or select agents

Nothing to report.

6. PRODUCTS:

- **Publications, conference papers, and presentations**
  Journal publications.

Nothing to report.

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

Nothing to report.

Other publications, conference papers and presentations.

Karunanayaka P, Mills-Huffnagle SL, Augusto CM, Cauffman A, Hazelton J, Kanekar S, Hobkirk A, Nyland JE. Disruption of default mode network and salience networks dynamics in acute traumatic pain states. Presented at the International Society for Magnetic Resonance in Medicine Annual Meeting (ISMRM), Toronto, ON, Canada, June 3-8, 2023.
Nyland JE, Janicki PK, Adhikary S, Giampetro D, Hazelton J, Staszak R, Mills-Huffnagle SL, Augusto CM, Sipe A, Cauffman A, Hobkirk A, Kanekar S, Karunanayaka P. Predicting pain chronification following traumatic injury. Presented at the Military Health Systems Research Symposium (MHSRS), Kissimmee, FL, August 14-17, 2023.

- **Website(s) or other Internet site(s)**

Nothing to report.

- **Technologies or techniques**

Nothing to report.

- **Inventions, patent applications, and/or licenses**

Nothing to report.

- **Other Products**

Nothing to report.

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

What individuals have worked on the project?

Name:	Jennifer Nyland
Project Role:	No Change
Name:	Aimee Cauffman
Project Role:	No Change
Name:	Prasanna Karunanayaka
Project Role:	No Change
Name:	Piotr Janicki
Project Role:	No Change
Name:	Ann Sipe
Project Role:	No Change

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

Jennifer Nyland

Pending grants now active:
Title: Evaluation of Inflammation in The Locus Coeruleus During Physical Withdrawal Symptoms and Cognitive Development in a Rat Model of Neonatal Opioid Withdrawal Syndrome (NOWS)
PD/PI: Mills-Huffnagle (Nyland: Sponsor)
Time Commitments: NA
Supporting Agency: NIH/NIDA F31 DA059237
Scientific Overlap: None

Jennifer Nyland (Continued)

Pending grants now active:
Title: Equipment Supplement for "Immune and neuroendocrine mediators of sex-differences in pain following traumatic burn injury"
PD/PI: Nyland
Time Commitments: NA
Supporting Agency: NIH/NIGMS F35 GM146774-2S01
Scientific Overlap: None

Aimee Cauffman
Nothing to report.

Piotr Janicki
Nothing to report.

Prasanna Karunanayaka
Nothing to report.

Ann Sipe
Nothing to report.

What other organizations were involved as partners?

Nothing to report.

8. SPECIAL REPORTING REQUIREMENTS

COLLABORATIVE AWARDS:

QUAD CHARTS:

9. APPENDICES: