Mapping of motor thalamic recording tracts using a combination of imaging, behavioral, and stimulation techniques.

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Background

The subnuclei of the motor thalamus are involved in motor control in diverse ways and are implicated to be affected in Parkinson's Disease (PD).

Thalamic Nucleus	**Input Region**	**Main Output Region**	**Behavior Encoding** (Vitek et al, 1994)	**Stimulation Response** (Vitek et al, 1996)
Nucleus ventralis anterior (VA)	Basal Ganglia	Pre-SMA and PMC	Active	No
Nucleus ventralis lateralis pars oralis (VLo)	Basal Ganglia	SMA and PMC	Active	No
Nucleus ventralis posterior lateralis pars oralis (VPLo)	Cerebellum	M1	Passive	Yes

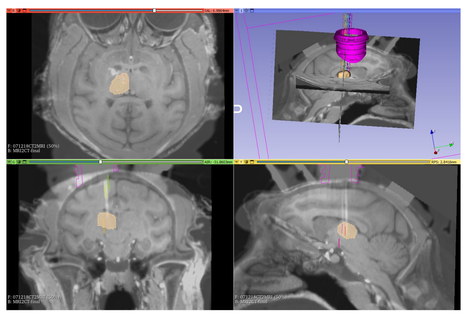
Experiments in our lab have used electrodes targeting the VA, VLo, and VPLo to record neural activity in the motor thalamus during different motor tasks. It is important to confirm that recordings were in these locations due to the difference in connections and behavior from one nucleus to the next.

The goal of this project was to determine the thalamic nuclei that were targeted by each recording electrode using a combination of imaging techniques, active movement, passive manipulation, tactile response, and microstimulation. This work will allow our lab to be able to say where prior recordings were performed in the thalamus.

Methods

Imaging Techniques

a. Using 3D slicer
   i. align an pre-implantation MRI and post-implantation CT scan
   ii. align 3D model of implant - 96 Channel Gray Matter Research Microdrive,
b. Merge standard brain atlas (Calabrese et al., 2015) with MRI images



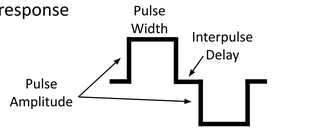
Neural Mapping

a. Measure depth of target along electrode tract in 3D slicer
b. Move recording electrode through the target, stopping at individual cells to determine responsiveness

Response Types				
None	Passive	Active	Tactile	Visual

Microstimulation

a. Determine stimulation threshold in the cortex (M1/PMC) that elicits a muscle response.
   i. Test low to high current until there is a muscle response
b. Stimulation Parameters
   i. Stimulation Frequency = 400 Hz
   ii. Pulse Width = 200us
   iii. Interpulse Delay = 100us
   iv. Pulse Duration = 200ms train
   v. Pulse Amplitude = Variable to determine the threshold for movement
c. Stimulate on thalamus electrodes with cortical levels as reference for parameter settings
   i. Distinguish between thalamic subnuclei



NIH NS037019 NS058945 NS077657 NS123109 NS117822

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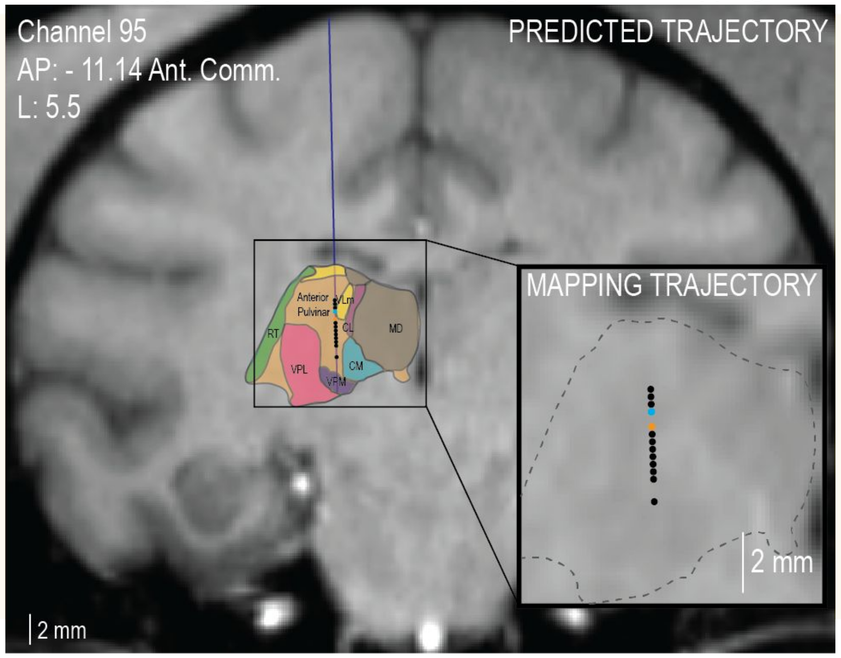
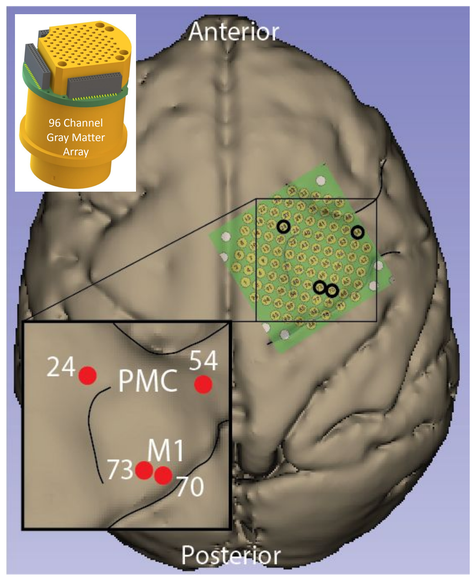
Results

Thalamic Mapping - Combination of Imaging and Neural Mapping



Most neurons found along channel 95 tract did not respond to any of the manipulation (black circles). A cell (blue circle) at 3.25mm into the thalamus responded to passive manipulation of the elbow and shoulder and a cell (orange circle) at 3.75mm into the thalamus responded to deep pressure in tricep and shoulder.

Microstimulation - Parameter Testing in Cortex



Channel 73 - M1			
Day 1	3.4mm	15mA	Pectoral contractions
Day 2	3.4mm	85uA	Bicep / Tricep contraction, moves arm forward
Day 3	3.4mm	60uA	Bicep / Tricep contraction, moves arm forward
Day 4	3.4mm	67uA	Bicep / Tricep contraction

Channel 70 - M1			
Day 1	3.2mm	70uA	Wrist movement and visible forearm twitch
Day 2	3.2mm	45uA	Subtle wrist inward rotation and forearm twitch
Day 3	3.2mm	50uA	Wrist movement, forearm contract

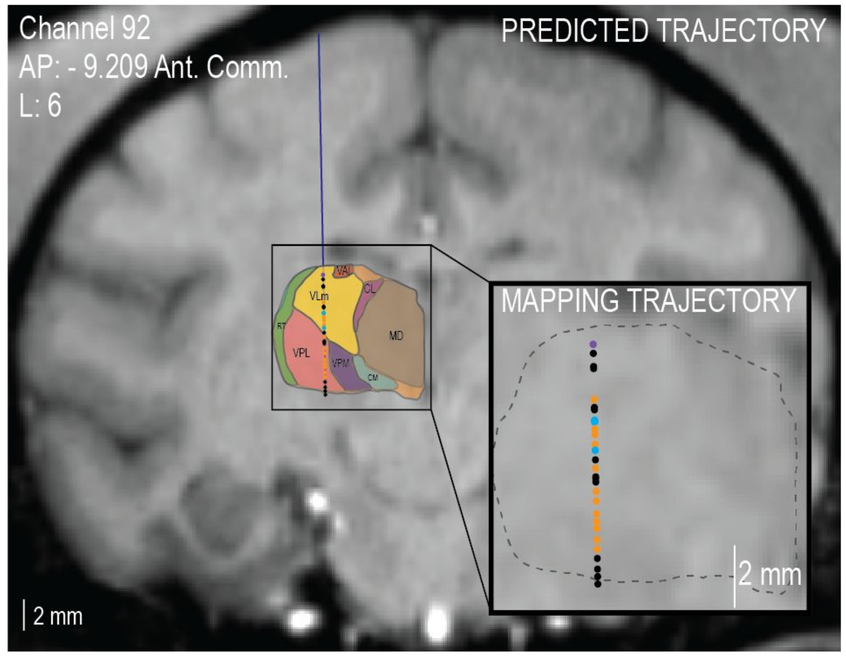
Channel 54 - PMC			
Day 1	4.5mm	75uA	Felt pulse/twitch in tricep (Passive shoulder ext, NOT tactile)
Day 2	4.5mm	75uA	Forearm/tricep contract

Channel 24 - PMC			
Day 1	3.5mm	60uA	Hamstring contraction(clear active movement of leg retraction, maybe deep pressure in hip)
Day 2	3.5mm	50uA	Hamstring/calf movement

We were able to elicit movements using microstimulation in both the motor cortex (M1) and the premotor cortex (PMC). Repeat testing days resulted in a decrease in the current needed to elicit a similar movement in the M1 until it reached a floor. PMC stimulation currents were similar on repeat test days to elicit the same type of movement.

Glossary

AP = anterior - posterior plane
CM = centromedian thalamus
CL = centrolateral thalamus
L = lateral plane
MD = mediodorsal thalamus
M1 = motor cortex
SMA = supplementary motor area
PMC = premotor cortex
VAl = ventral lateral thalamus, lateral part
VLm = ventral lateral thalamus, medial part
VPL = ventral posterolateral thalamus
VPM = ventral posteromedial thalamus



Most neurons found along channel 92 tract responded to tactile stimuli (orange circles) on the face, torso and leg from dorsal to ventral. There were a couple cells that responded to passive movement (blue circles) and the very first cell in the tract responded to visual stimuli (purple circle).

Conclusions

Thalamic Mapping

- Channels 95 and 92 are posterior of the motor thalamus, with 92 being potential on the border.
- The standard atlas is a good guideline to use when predicting thalamic nuclei, but confirmation with mapping is still ideal.

Microstimulation Testing

- M1 and PMC microstimulation is effective at eliciting motor movements, with M1 requiring less current than PMC.
- The amount of current required for M1 and PMC is slightly higher than expected.
  - Likely due to current loss during stimulation through the GMA system.
  - Potentially due to stimulation in PD state.

Future Directions

1. Continue to map electrodes moving more anterior to channel 92, which should be in the motor thalamus. We expect to see cells along these tracts will respond to passive or active movement, not to tactile or visual stimuli.
2. Microstimulation of all thalamic channels that are thought to be in the motor thalamus based on neural mapping results. This will allow us to determine a border between VPLo and VA/VLo.
3. Stimulation lesions + histology will further confirm electrode locations.

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cFOS expression in hippocampal CA2 with cerebellar vermis activation

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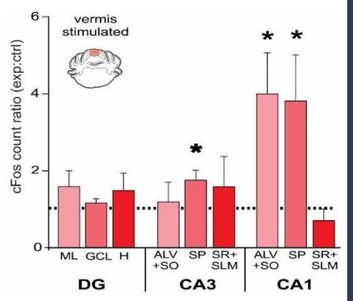
Introduction

- The hippocampus, a structure synonymous with memory in the brain, has been shown to be influenced by the cerebellum, which is generally associated with movement but also has roles in cognitive functions.

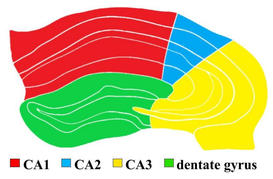
Prior work in the KM lab has demonstrated a functional connection between the cerebellum and the hippocampus.[1]

- One way to study this connection is by analyzing cFOS activation in the different layers/subregions of the hippocampus (e.g. CA1 and CA3)
- This previous work (right) has shown the cerebellum can affect cFOS expression in multiple layers/subregions of the hippocampus, but little is currently known about the CA2 region's reaction to cerebellar influence.



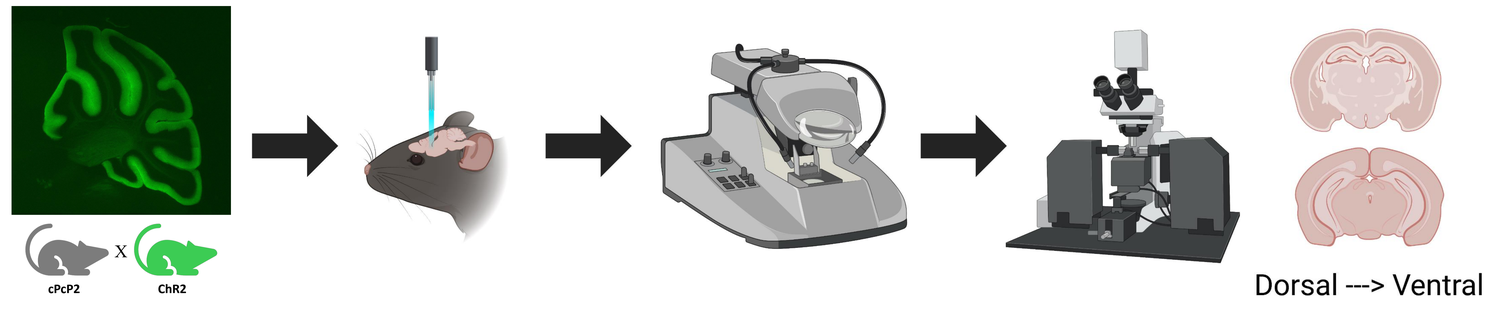
In this experiment, we are looking at if CA2 cells are activated or not with cerebellar stimulation via looking at cFOS expression.

- Results from this experiment could both lead to better understanding of the connection between the cerebellum and hippocampus and increase interest in the CA2 region of the hippocampus with regards to this circuit.

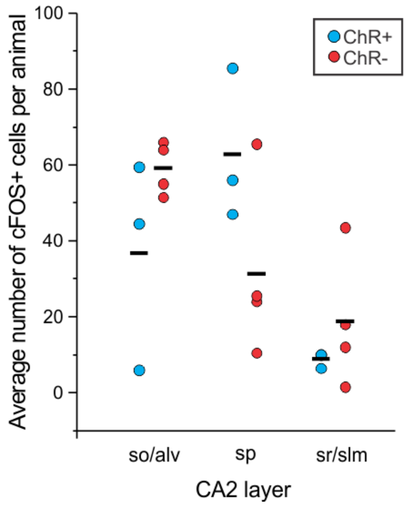


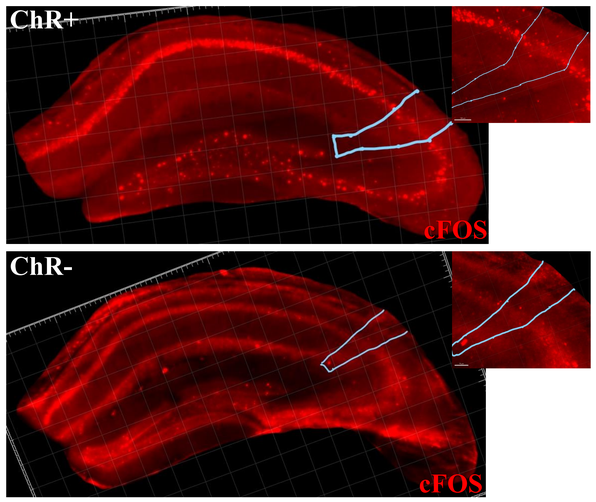
Methods

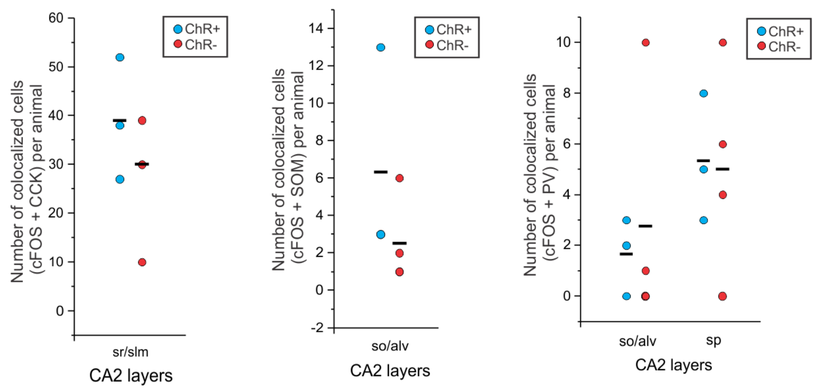
1. Mice were bred to express channelrhodopsin (ChR) in Purkinje cells. ChR negative littermates were used as controls.
2. Animals were optogenetically stimulated with blue light for a period of 1 hour (7Hz, 50 ms on 100 ms off) in a home cage environment.
3. The mouse brains were perfused, extracted, and sliced at 50um coronally using a vibratome.
4. Brain tissue was mounted on microscope slides and imaged as Z-stacks at 20x with a Zeiss microscope for tiling and stitching of each image.
5. The images were visualized in the Imaris program for counting cells that were cFOS+ and/or CCK+/PV+/SOM+ in CA2.

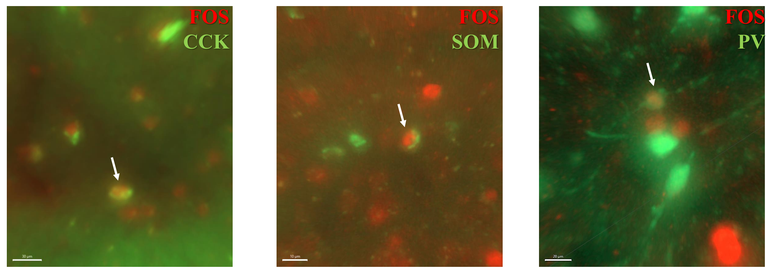


Preliminary Results









Amount of cFOS + and interneurons colocalized with cFOS in CA2 layers per animal. Graphs represent the summed total of all cells in 5 slices per animal in each CA2 layer. For all, blue represents cells per layer in ChR+ mice (N=3) and red indicates cells in ChR- littermates (N=4). Images are from representative slices to show relative cFOS differences (above) and colocalization examples (right). ***(above, left)*:** Average number of cFOS + cells per animal by CA2 layer ***(above, middle):*** Representative images of cFOS in example mice. Note CA2 region outlined in blue and in the zoomed in image. ***(top, right):*** Colocalized cells per CA2 layer (i.e. cFOS + interneuron expression). ***(bottom, right*):** Example of colocalized cells [cFOS (in red using AF568 filter) + subset of interneuron (in green using EGFP filter)]. ***Abbreviations:*** cholecystokinin- CCK; Somatostatin- SOM; Parvalbumin- PV; So/alv- stratum oriens/ alveus; sp- stratum paramidale; sr/slm- stratum radiatum/ stratum lacunosum moleculare.

Discussion

Results so far:

- cFOS may be increased in opsin positive animals specifically in the SP layer of CA2
- Colocalization results (CCK, SOM) may suggest recruitment of specific hippocampal interneurons with cerebellar stimulation
- Lots of animal-to-animal variability noted—this is a caveat to potential results

Future directions:

- Adding more animals
- Look at cFOS across A/P axis

Acknowledgements

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What Makes Enzymes Faster?

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Project supported by the Louis Stokes North Star STEM Alliance

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Introduction

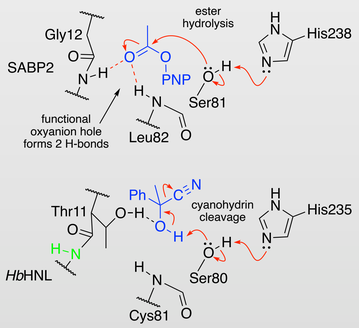
Enzymes are biological catalysts that accelerate chemical reactions. Understanding enzyme efficiency is imperative in the process of making chemistry more sustainable because it can replace harmful non-sustainable catalysts.
Exploring how enzymes evolve and adopt different functions can help us understand how to manipulate them.

- **Hydroxynitrile Lyase (HNL)**: responsible for the synthesis of cyanohydrins) evolved from esterase
- **Esterase (SABP2)**: catalyzes hydrolysis of esters.
- HNL evolved from SABPB2 approximately 100 million years ago.

That evolutionary process can be understood by giving HNL the functions of SABP2 through mutations of HNL. **Learning what mutations makes HNL an efficient esterase, and what extends its functions contribute to the general knowledge about how enzymes work and can lead to more sustainable scientific practice.**

Different versions of HNL with different mutations have been found to lead to efficient enzymes with great esterase functions. HNL3V has poor esterase activity, however substituting asparagine 104 with alanine improves catalytic activity (HNL3VN104A). pH dependence of the enzymes may play a part in this difference in catalytic activity.

Hypothesis and Goals



HNL3V has significantly lower esterase activity that its variant HNL3VN104A. The only difference between these enzymes is a mutation in the active site. How could this effect the catalytic activity of the enzyme?

Hypothesis: These enzymes may have different pH dependence.

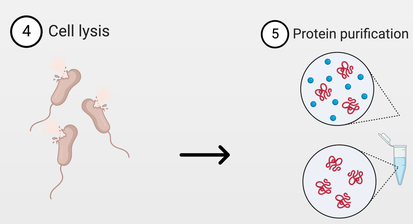
Goals: To grow bacteria, purify the enzymes and measure their catalytic activity at different pH

Methods

Protein Expression and Purification

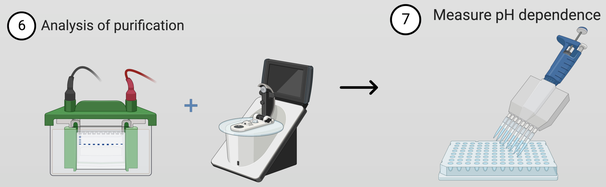
Plasmid were obtained that could grow these specific enzymes: HNL3V and HNL3VN104A, SABP2 and SABP2A104N.

Electroporation creates small pores in the cell wall allowing for plasmids to enter. Cells that grow on ampicillin-containing plates have acquired the plasmid. These bacteria are grown in larger cultures. Adding IPTG turns on overexpression of the target protein.



CELL LYSIS: Cell Lysis is the process of breaking open the cells. Freezing the samples along with sonification causes cell lysis. Sonification agitates the cell wall with sound bursts. When the cell breaks, protein is released into the supernatant.

PROTEIN PURIFICATION: The supernatant that contains the protein is incubated with His-tagged resin which binds to the target protein and retains it in a column while the other content is filtered out. Elution buffer replaces the bind of the histidine tag, attaches to the target protein, allowing it to be filtered out.



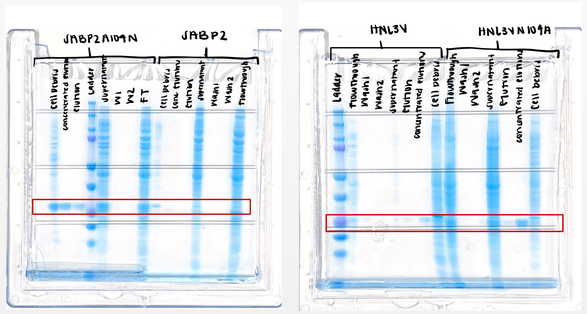
SDS-PAGE separates protein based on size. Since the molecular weight of the desired protein is shown on the gel (by size), protein location is indicated by that signal. Nanodrop measures the absorbance as well the ratio of protein to DNA. Concentration can be obtained from the absorbance using Beer's Law.

pH Dependence is determined by measuring the rate of a reaction while changing the pH of the reaction. Finding the isosbestic point is necessary to ensure that the changing pH does not affect absorbance of the reaction product, p-nitrophenol.

Results

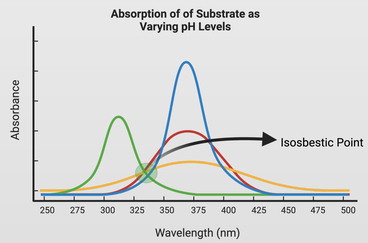
Protein was correctly purified; however, the pH dependence was not determined because protocol has not yet been perfected and it yielded too much error. Instead, the expected pH dependency results will be discussed.

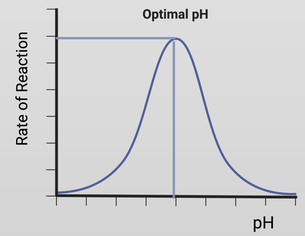
Protein Expression and Purification Results



The SDS page checks that the enzyme was made. The red marked area on the gel is where the protein should be located. The volumes that represent the enzyme (the concentrated elutions) appear to be the most purified, despite slight contamination.

Catalytic Activity Expected Results





In order to measure pH dependence, the formation of product is measured at its isosbestic point. At this isosbestic point the absorbance of the product does not vary with pH. At the determined isosbestic point, the pH dependence measurement is more accurate.

Conclusion/Discussion

In this project, one of two goals is complete: growing and purifying protein. Once the enzyme was obtained, isosbestic point determination as well as the pH dependence was attempted. While no tangible results were obtained, questions and confusions surrounding the protocol will contribute to eventual success to measuring pH dependence.

The research questions: "What makes Enzymes fast?" is a significant question that was never going to be fully answered with results but would have helped with the understanding of how pH comes into play regarding enzyme activity. The hypothesis was created based on the known fact that pH can easily change enzymatic activity, which could explain why only one mutation could have such a drastic effect of catalytic activity.

It has also been found in other studies that this change lowers the pKa of the active sites on the enzymes. This means that the protonated version of histidine is unstable, and exists more as a base, more capable of catalyzing hydrolysis of esters and consequently having more enzyme activity.

The hypothesis cannot be ruled as true or not, as there is no experimental data. However, changes to the protocol can be suggested to improve results in the future. The most significant problems to address would be related to repeating the experiment multiple times to determine the best concentration of each component in the reaction (buffer, enzyme, and substrate). Another important factor to consider is the speed of which assays are done, and how to make sure the reaction does not dully hydrolyze before it can be measured

Despite these edits, understanding the base impact of pH regarding measuring enzymatic activity is mandatory to learn more about the manipulation of these enzymes to eventual lead to more sustainable scientific practices.

Acknowledgements and References

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A guideline to optimize neuroimaging data collection using mobile fNIRS

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Background

- Functional near-infrared spectroscopy (fNIRS) is a relatively new neuroimaging tool[1]
  - Sources/Detectors[2]
  - Hemoglobin/Deoxyhemoglobin[2]
- Has numerous benefits over traditional technology (MRI and EEG)[3,4]
  - Portable and mobile
  - Allows for more naturalistic experiments
  - More comfortable and child-friendly

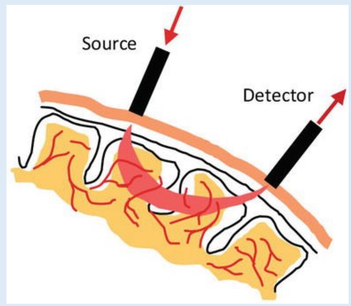

Figure 1: fNIRS Source and Detector (NIRx medical technologies)

- Because the technology is newer, however, there is less research on how to optimize data collection using fNIRS relative to other neuroimaging tools[5]
- Our objective was to develop a guideline to attain quality neuroimaging data using NIRSport2 - a new mobile fNIRS device

Methodology

Cap Sizing

1. Measure participants head from Nz to Iz (Figure 1)
2. Take 10% of the measurement and mark that length above the eyebrows and the base of the skull
3. Find the circumference of the subject's head using those 10% markers
4. Measure from the RPA through through to the LPA
5. Using the measure of the circumference, find the appropriate cap for the participant (if odd number use the previous even number)
6. Apply cap onto subject's head to test if it truly fits

Cap Preparation

1. Apply appropriate optodes and spring holder top to mesh cap that has a premade montage design on it
2. Place finished cap onto subject's head and measure the distance from the Nz to the Cz and the Cz to the Iz to make sure that both measurements are equal
3. Do the same for the points at the ears and the Cz
4. Apply black shower cap atop of everything else to reduce environmental noise

Connecting fNIRS Device

1. Connect NIRSport 2 wirelessly to a computer and open Aurora–a data acquisition software[6]
2. Calibrate Signal
3. Monitor signal levels, noise tables, and brain montage
4. Record data

Results

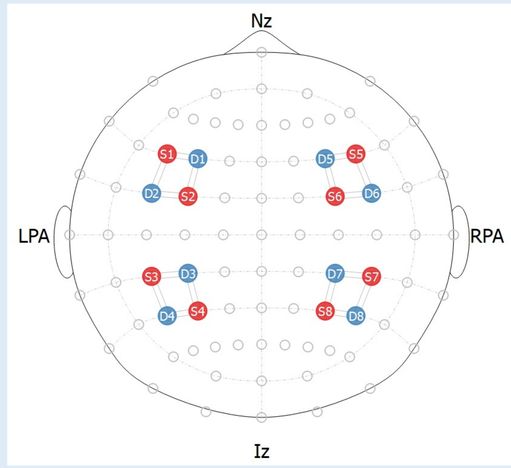

Figure 2: Montage and Mapping of the fNIRS optodes as they are arranged on a mesh cap. The regions closest to the front of the head sit atop the dorsolateral prefrontal cortex (DLPFC) while the other two are placed above the temporoparietal junction (TPJ).

Participant 1 **Participant 2**

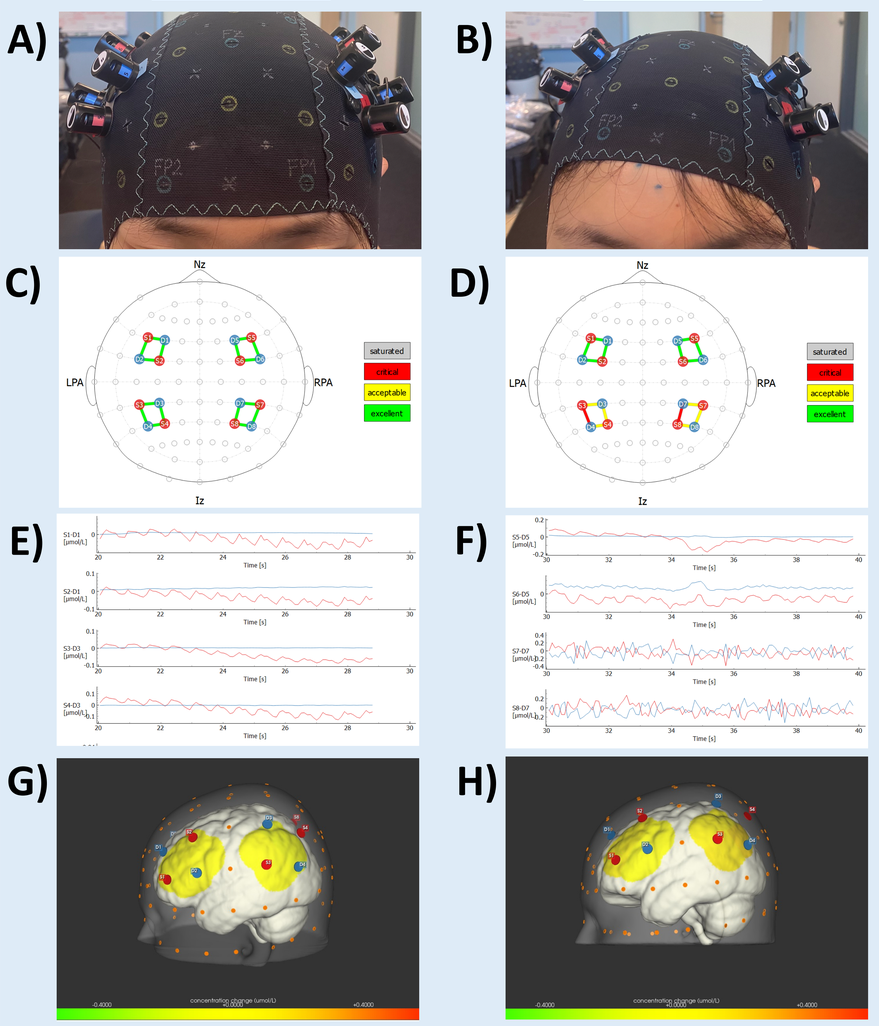

Figure 3: Depictions of what are considered both "good" (left) and "bad" (bad) demonstrations of a fNIRS cap set up (A-B), signal calibration (C-D), noise table (E-F), and brain model (G-H). The cap set up images are independent of the latter figures. In terms of the last six figures, the two columns of images correspond to data from two different participants.

Guidelines

- Montage designs should be designed to correspond to the region of the brain being studied in an experiment
  - Ex: Cognitive Flexibility → DLPFC and TPJ[7]
- Mesh cap should fit well to the participants head and be centered between the eyebrows
- In terms of signal calibration, green, yellow, and red denote good, acceptable, and bad, respectively
- The noise tables also show whether a signal is good or not; the blue line should be relatively straight while the red should fluctuate
  - Blue line → deoxyhemoglobin
  - Red line → hemoglobin
- For brain montages, the respective regions should appear as a light-yellow color

Conclusion

- The purpose of this study was to develop a method on how to complete a basic experimental paradigm with fNIRS
- Better methods → good quality data
- Next Steps:
  - Conduct an experimental paradigm using these basic steps
  - Complete a new study that investigates fNIRS and different hair textures

References

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Acknowledgements

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A demonstration on how to use and collect data with fNIRS to increase the likelihood of obtaining quality data

Elham Osman and Dr. Ka Ip
Institute of Child Development, University of Minnesota – Twin Cities

Background

- Functional near-infrared spectroscopy (fNIRS) is a relatively new neuroimaging tool[1]
  - Sources/Detectors[2]
  - Hemoglobin/Deoxyhemoglobin[2]
- Has numerous benefits over traditional technology (magnetic resonance imaging and electroencephalography)[3,4]
  - Portable
  - Allows for more naturalistic experiments
  - More comfortable
- Because the technology is newer, however, there are less published articles on how to successfully conduct studies with fNIRS as compared to MRI and EEG[5]
- Due to this gap, our main goal was to successfully develop a method to attain quality data while using fNIRS

Methodology

Cap Sizing

1. Measure participants head from Nz to Iz (Figure 1)
2. Take 10% of the measurement and mark that length above the eyebrows and the base of the skull
3. Find the circumference of the subject's head using those 10% markers
4. Measure from the RPA through through to the LPA
5. Using the measure of the circumference, find the appropriate cap for the participant (if odd number use the previous even number)
6. Apply cap onto subject's head to test if it truly fits

Cap Preparation

1. Apply appropriate optodes and spring holder top to mesh cap that has a premade montage design on it
2. Place finished cap onto subject's head and measure the distance from the Nz to the Cz and the Cz to the Iz to make sure that both measurements are equal
3. Do the same for the points at the ears and the Cz
4. Apply black shower cap atop of everything else

Connecting fNIRS Device

1. Connect NIRSport 2 wirelessly to a computer and open Aurora[6]
2. Calibrate Signal
3. Monitor signal levels, noise tables, and brain montage
4. Record data

Results

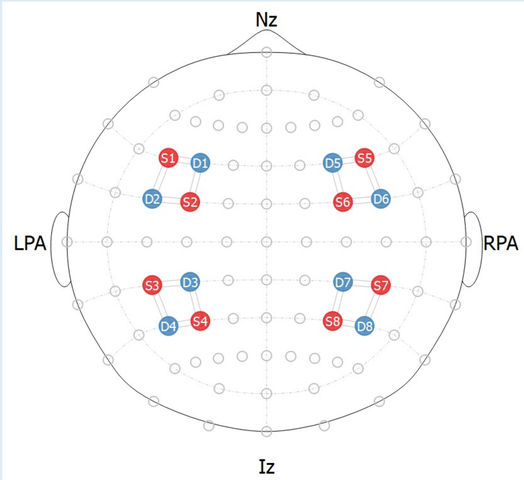

Figure 1: Mapping of the fNIRS optodes as they are arranged on a mesh cap. The regions closest to the front of the head sit atop the dorsolateral prefrontal cortex (DLPFC) while the other two are placed above the temporoparietal junction (TPJ).

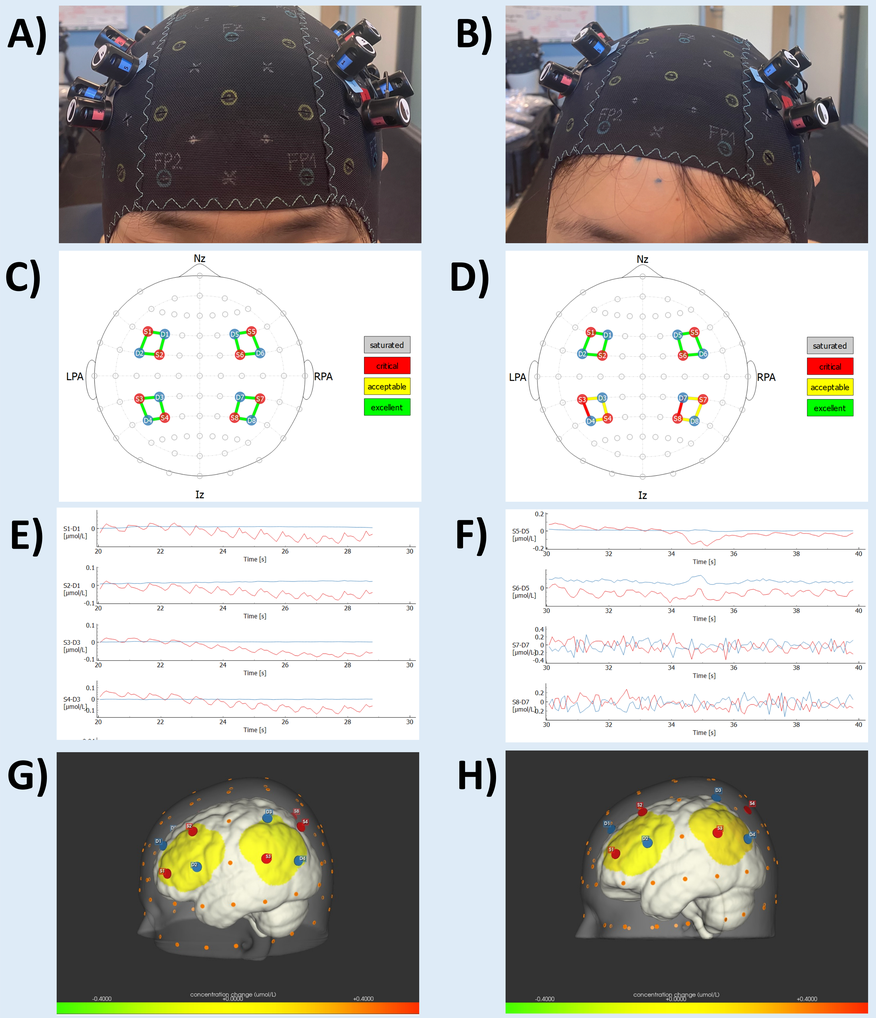

Figure 2: Depictions of what are considered both "good" (left) and "bad" (bad) demonstrations of a fNIRS cap set up (A-B), signal calibration (C-D), noise table (E-F), and brain model (G-H). The cap set up images are independent of the latter figures. In terms of the last six figures, the two columns of images correspond to data from two different participants.

Discussion

- Montage designs should be designed to correspond to the region of the brain being studied in an experiment
  - Ex: Cognitive Flexibility → DLPFC and TPJ[7]
- Mesh cap should fit well to the participants head and be centered between the eyebrows
- In terms of signal calibration, green, yellow, and red denote good, acceptable, and bad, respectively
- The noise tables also show whether a signal is good or not; the blue line should be relatively straight while the red should fluctuate
  - Blue line → deoxyhemoglobin
  - Red line → hemoglobin
- For brain montages, the respective regions should appear as a light-yellow color

Conclusion

- The purpose of this study was to develop a method on how to complete a basic experimental paradigm with fNIRS
- Better methods → good quality data
- Next Steps:
  - Conduct an experimental paradigm using these basic steps
  - Complete a new study that investigates fNIRS and different hair textures

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Do Spontaneous Activity Patterns Predict Functional Organization in Primate V1?

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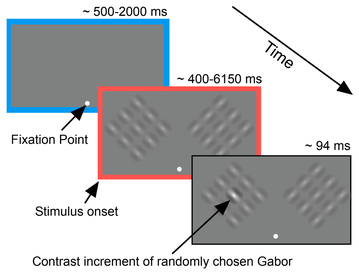
University of Minnesota Twin Cities, Department of Neuroscience

Introduction

The processing of sensory information in the cerebral cortex is thought to be guided by close groups of neurons over a scale of hundreds of microns that exhibit similar functional characteristics. These neurons demonstrate a modular organization of the brain and can guide approaches to understanding why spatial patters of coactivation in the brain may look they way they do. New studies propose that patterns of brain activation caused by stimulation are due to the inherent properties of the cortex and the relationship between the different regions of neuron groupings. To investigate this hypothesis, we compared the brains activity patterns during visual stimulation with those observed in spontaneous activity before any sort of visual input.

Methodology

- Flavoprotein fluorescence imaging is a neuroimaging technique that utilizes the unique optical properties of flavoproteins, which emit fluorescence when excited by light, to provide a non-invasive visualization of neuronal activity
- Principal Component Analysis (PCA) is a mathematical method commonly used to reduce the complexity of a dataset while preserving essential patterns and correlations.
- PCA was applied to analyze the acquired imaging data transforming the fluorescence intensity values into a new set of orthogonal components, revealing the correlation between spontaneous activity in the primary visual cortex and the modular organization of the visual system for processing specific visual stimuli.



Monkeys performed a challenging visual task in which they were required to maintain gaze upon a central fixation point until a small contrast increment occurred at one of 18 patches. All patches were of the same orientation and drifted in phase with each other. Data used for this paper is from spontaneous activity (blue, prior to stimulus presentation) and immediately after stimulus presentation, but prior to the behaviorally relevant contrast change

Visualization of how spatial distribution of primary PCA components change as a function of Gabor location.

Image Processing

Utilize flavoprotein fluorescence imaging to capture neural responses in the Monkey brain

↓

Employ a high-resolution imaging system to record fluorescence signals in the cortex

↓

Preprocess and correct for motion artifacts and image distortions in the acquired fluorescence images using custom scripts in MATLAB,

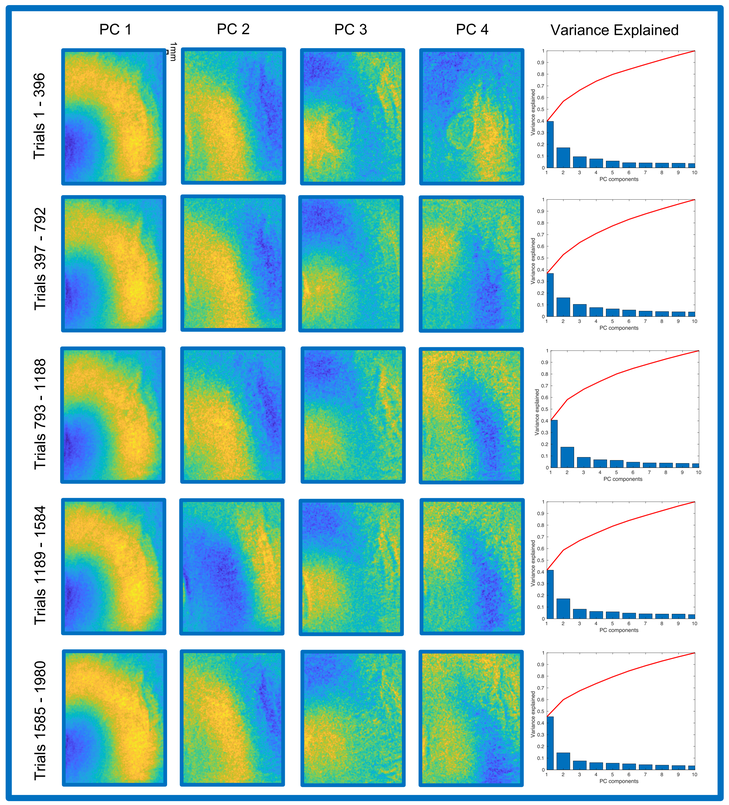
↓

Apply PCA to the processed fluorescence imaging data using MATLAB's PCA function from the Machine Learning and Robot Vision toolbox

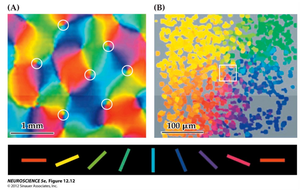
↓

Visualize variation in activity captured by the principal components over time and identify patterns or clusters of neural activity

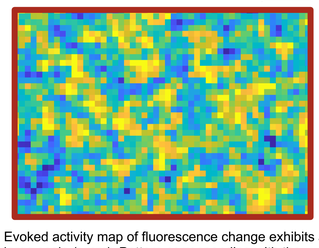
Results



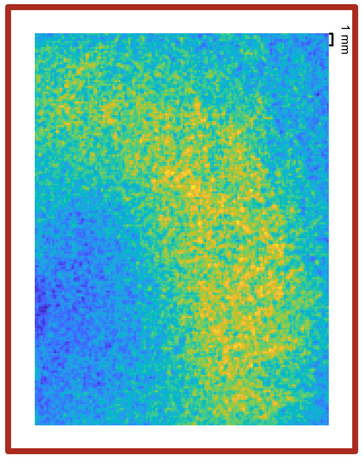
Decomposition of spontaneous activity according to component (PC). The first four components, arranged in ascending order from left to right, along with their respective proportions of variance explained, depicted in the rightmost plot. PC decomposition was carried out for five groups of 396 trials shown in the rows above, resulting in a total of 1980 trials.



Functional imaging illustrating modularity of V1 based on orientation preference.

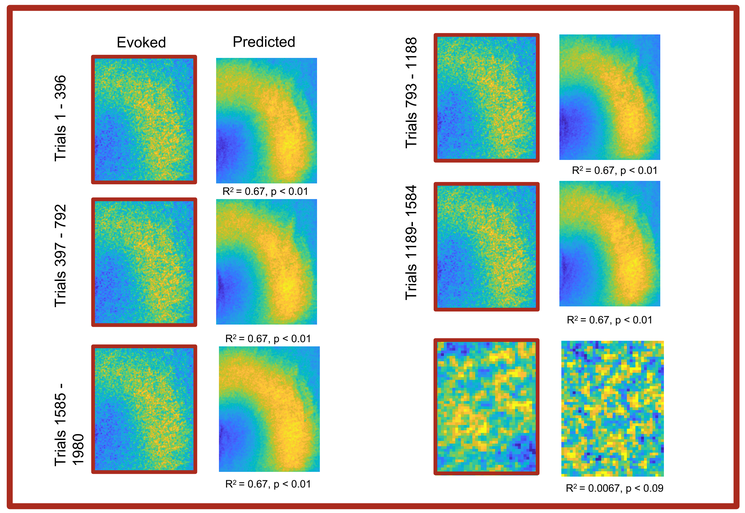


Evoked activity map of fluorescence change exhibits a large scale (>mm). Pattern corresponding with the retinotopic extent of the stimuli (above) and a fine scale (<mm) pattern of orientation selectivity across V1.



Averaged V1 evoked activity map of fluorescence change exhibits a distinct patchiness pattern indicative of the well-known lateral spread of orientation selectivity of the V1 region.

Results



Simple linear regression was utilized to evaluate the predictability of evoked activity using the first four principal components of spontaneous activity. Each of the five trial groups underwent a comparison between the average evoked activity on the left and the corresponding predicted evoked activity on the right. Additionally, in the final sub-plot, we concentrated on a more closely examined subsection of average evoked activity from trial group 1, which better reveals V1 orientation selectivity. Once again, we compared this with the predicted evoked activity using the principal components of spontaneous activity.

Conclusions

- Using flavoprotein autofluorescence measurements of the evoked activity from an array of stimuli of the same orientation, we are able to visualize patterns on functional organization in area V1 with respect to retinotopic position (on the order of mm) and orientation preference (on the sub-mm scale).
- Spontaneous activity is spatially organized and low-dimensional with a few principal components dominating.
- Linear sums of these principal components are consistently able to explain activity variations over the scale of mm (largely reflecting the retinotopy), but less able to explain the orientation specific pattern of activation we observe.
- The results suggest that the spatial scale over which intrinsic covariance constrains evoked activity is on the order of millimeters and is not sufficient to explain finer scale functional organization, such as orientation columns.

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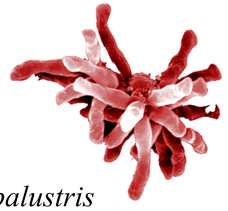
Phenotyping Electron Donor Variants for Nitrogen Fixation

Ezra Albright, Nathan M. Lewis, Kathryn R. Fixen

University of Minnesota

Introduction



Nitrogen Fixation

- Process where molecular Nitrogen (N_2) is converted into Ammonia (NH_3)

Haber-Bosch process vs *Rhodopseudomonas palustris*

- Fossil Fuel dependent
- High Temp
- CH4 In

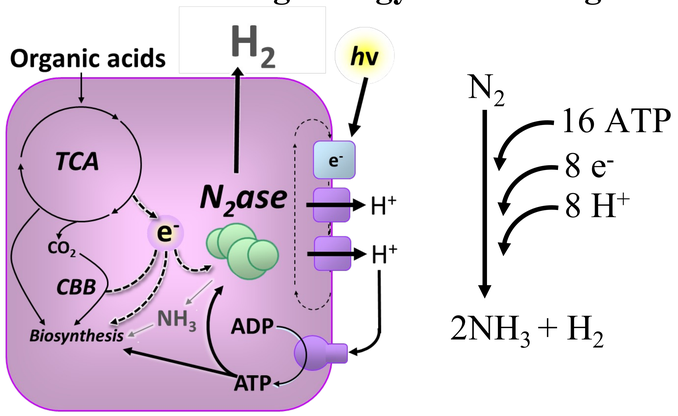
- Sunlight
- Renewable
- Creates valuable products

→ $2NH_3 + H_2$

Experimental Goals

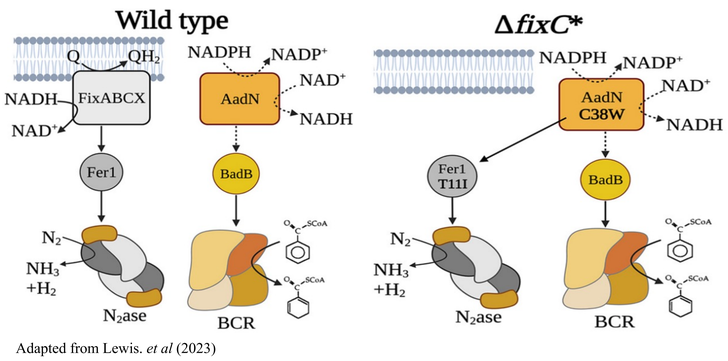
1. To explore how different variants of an electron transfer pathway effect growth
2. To determine what factors assist with growth in those specific variants
3. How that growth is affected in different media

Rhodopseudomonas palustris Catalyzes Nitrogen Fixation Using Energy from Sunlight



Adapted from McKinlay & Harwood (2010)

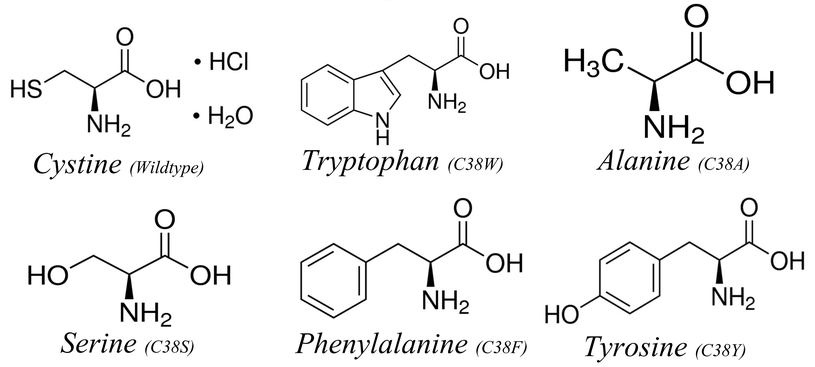
Mutations in AadN Form an Alternative Electron Transfer Pathway for Nitrogenase



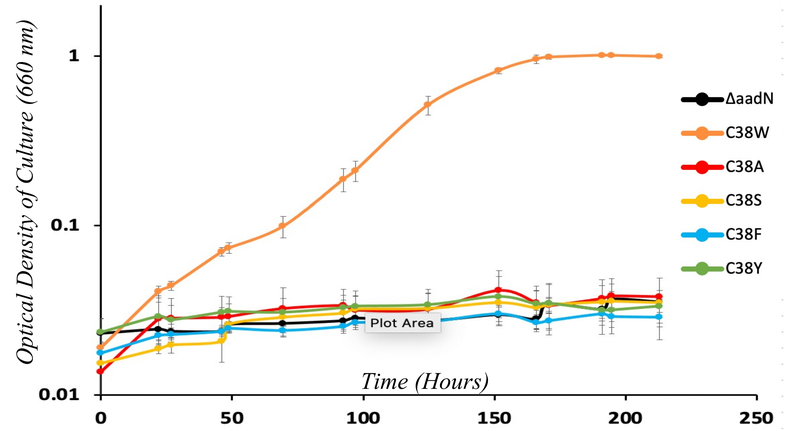
Adapted from Lewis. *et al* (2023)

Why C38W?

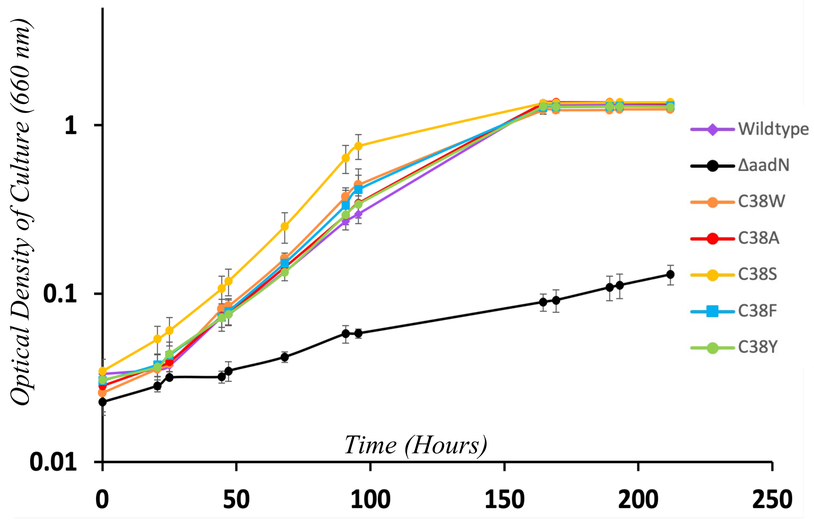
Characterizing AadN Variants



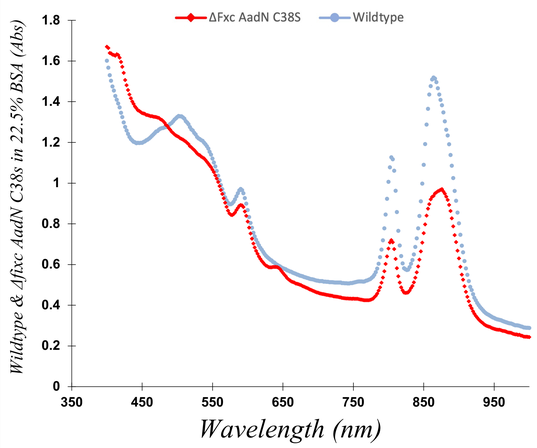
Only the AadN C38W Variant Can Support Electron Transfer for Nitrogen Fixation



All AadN Variants Support Electron Transfer for Benzoate Degredation



A Deeper Look at C38S' Color



Wildtype C38S

- Noticeable differences between the wildtype and C38S around ~475nm and lower peaks at ~800nm and ~900nm

Future Directions

- Why does Tryptophan work as opposed to other amino acid structures?
- What is the biochemical activity of AadN?
- Why does C38S differ in color so much and does it affect photosynthesis?
- Is the C38S causing a stress response in *R. palustris* and if so, it can help us understand more about what is going on

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Acknowledgements

FIXEN LAB
A RESEARCH LAB AT THE U OF MN

College of Biological Sciences

Special thanks to all the members of the Fixen Lab that helped set me up & guide me through my work this summer especially Kathryn R. Fixen Nathan M. Lewis

U.S. DEPARTMENT OF ENERGY

Minnesota's Louis Stokes Alliance for Minority Participation
MnDRIVE

The Effects of Setting on Tic Frequency in Individuals with Tic Disorders

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University of Minnesota, Department of Psychiatry & Behavioral Sciences

Introduction

- **Tics are sudden and involuntary movements or sounds that are done repeatedly.** While these movements are involuntary, they can be voluntarily inhibited for short periods of time. Tics can be classified as complex or simple.
- **Simple motor tics** can involve a few body parts, while **complex motor tics** can involve multiple body parts, have a pattern, or convey societal meaning, such as obscene gestures.
- **Simple vocal tics** can manifest as non-speech like noises (grunts, sniffs, coughs, etc.). **Complex vocal tics** can convey societal meaning or involve whole words or phrases.
- Tic disorders are typically assessed by a clinician in an assessment interview. Diagnosis and ratings of symptom severity are based on the patient's self- and/or parent-reported clinical history and clinician observations of the patient during the visit.
- **Tics can be reactive to context, such that the symptoms clinicians observe in clinical visits may differ from those experienced in daily life.** The extent to which tic expression differs between clinical assessments and settings in which the patient is alone has not been studied previously.

Study Aim: This study aims to assess how much tic frequency changes from a clinical interview to a different environment where a clinician is not present.

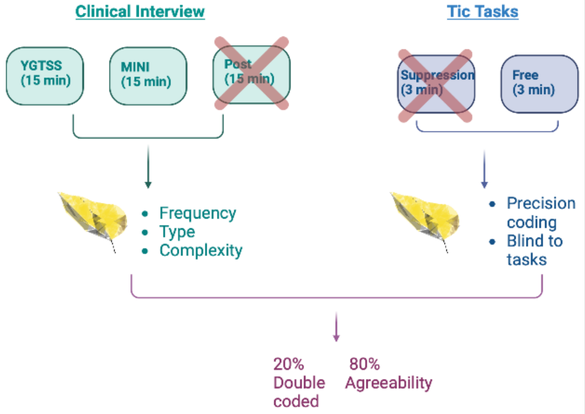
Hypothesis

We propose that the frequency of tics during clinical interviews (YGTSS & MINI) are lower than the frequency of tics during the Free Tic Task.

Methodology

Participants were selected from an ongoing **CBIT** (Cognitive Behavior Intervention Therapy) study. Pre- and post-treatment measures included:

- **Yale Global Tic Severity Scale** (**YGTSS):** The current standard , semi-structured interview for assessing tic disorders)
- **MINI-5:** Semi-structured interview assessing DSM-5 psychiatric diagnoses.
- **Tic Tasks:** Recorded using a go-pro in a lab room, where participants were alone and were asked to tic freely and then suppress their tics. Tasks are aimed to assess tic frequency which is measured by **tics per minute**.



Participant Characteristics

- 80% of participants identify as white
- 50% identify as male
- 50% identify as female
- Average age: 14.7 years
- **Average YGTSS severity score: 22.5**

Participant	Race	Sex	Gender	Age	YGTSS Severity Score
1	White	Female	Female	17	19
2	White	Male	Male	13	25
3	White	Female	Female	18	12
4	White	Male	Male	14	38
5	White	Male	Male	15	19
6	White	Female	Female	13	18
7	White	Female	Female	16	20
8	More than one race	Male	Male	14	29
9	White	Female	Female	16	25
10	N/A	Male	Male	11	20

Figure 2: This figure depicts the demographics of the participants by race, sex, gender, age, and YGTSS severity score

Results

- Our results indicate that there is a **large discrepancy between the frequency of tics during clinical assessments and the Free Tic Task, such that tic frequency is higher during the Free Tic Task.**
- Participants had an average of:
  - **120% difference** between the **Free Tic Task** and the **YGTSS** interview
  - **132% difference** between the **Free Tic Task** and the **MINI** interview
  - **20% difference** between the **YGTSS** and **MINI** interview
  - **75% difference** between the **clinical interview** and the **Free Tic Task**
- **Overall, the frequency of tics is higher for the Free Tic Task than the YGTSS and MINI interviews.**

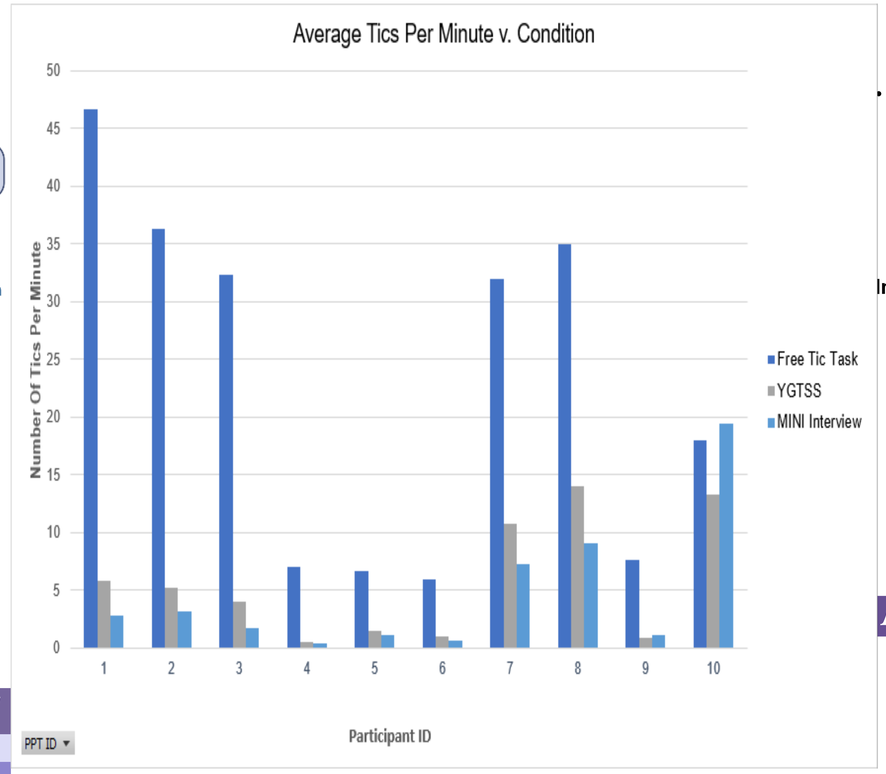


Figure 2: This figure depicts the difference of average tics per minute between each condition for each participant. There are **three trends** in the data, large differences, small differences, and an outlier. We ran paired t tests and found:
Free Tic Task & YGTSS: t = 4.07, df = 9, p < .01
Free Tic Task & MINI: t = 3.73, df = 9, p < .01
YGTSS & MINI: t = 1.077, df = 9, p = .3095.

Conclusion

- It is well known that tics manifest differently within different settings. However, **the disproportionately low frequency of tics during the clinical interviews indicate that during clinical assessments, clinicians may not get an accurate representation of tic frequency outside of a clinical setting.** This indicates that better assessment tools need to be developed in order to accurately diagnosis and subsequently treat patients and monitor the effects of treatments.
- These results indicate that when conducting research on individuals with tic disorders, comparisons of tic expression during clinical assessments versus settings **will be more representative of daily life in larger samples.**

Implications for future research

- Reliability of self reporting
- Inclusion of video-based measures such as the Tic Task in clinical trials
- Realistic treatments
- How conditions where symptoms may differ by context should be examined (Autism, OCD, etc.)

Acknowledgements

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Research Evaluating Vagal Excitation and Anatomical Linkages (REVEAL) on the effect of Vagal Nerve Stimulation

Insia Kizilbash[1], John Osborn[2], Miguel Anselmo[3], Emma Lee[1], Manda L. Keller-Ross[1,3]

[1]Division of Physical Therapy, [2]Division of Surgery, [3]Division of Rehabilitation Science, University of Minnesota

Minnesota's Louis Stokes Alliance for Minority Participation – MnDRIVE

University of Minnesota – Driven to Discover℠

Introduction

- The autonomic nervous system (ANS) maintains homeostatic control in the body and is involved in regulation of temperature, cardiovascular, metabolic and inflammatory systems.[2,3]
- The ANS consists of sympathetic and parasympathetic activity that work interdependently.
- Vagus Nerve is the primary nerve involved in parasympathetic function.
- Vagus Nerve Stimulation (VNS) is approved to treat epilepsy (1997) and depression (2005), however, its influence on physiological systems is unknown.[1]
- Sympathetic nervous system (SNS) activity increases with disease; high SNS can lead to end organ damage.
- How vagus nerve stimulation influences SNS activity is unclear, but could lead to novel and innovative therapies.

Purpose & Hypothesis

Purpose: Use a variation of VNS stimulation parameters to determine acute and chronic effects of VNS on sympathetic function.

Hypothesis: We hypothesize that VNS stimulation will reduce sympathetic activity at rest and during ANS perturbations.

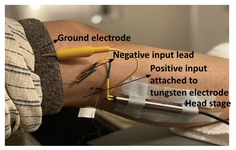
Methods

General Overview of REVEAL Clinical Trial

- The REVEAL study is a multi-site, single-blinded, crossover study.
- The target sample size is 144 participants of 18 years of age or older.
- Six parameters of acute and chronic VNS are received by participants in a randomized order.
- Measuring various parameters throughout help us study in particular the cardiac and autonomic systems in relation to VNS.

Specific to the Aims of this study

- Participants will attend two visits with continuous monitoring of BP, sympathetic activity, and HR using parameters of VNS during rest, exercise, and head up tilt.



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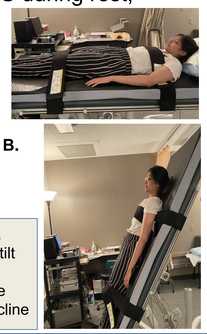

Figure 3. Head up tilt (right). ***A)*** Supine ***B)*** 60° incline

Methods

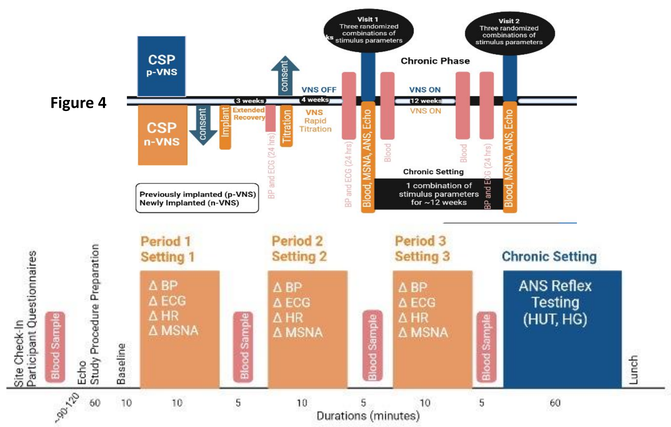

Figure 5a

Visit 1 Timeline

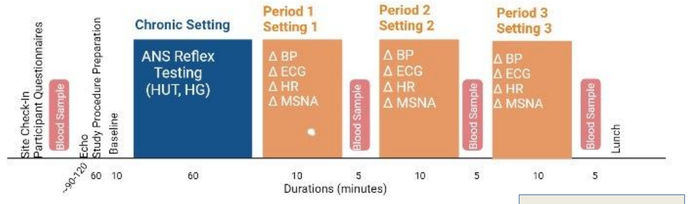

Figure 5b

Visit 2 Timeline

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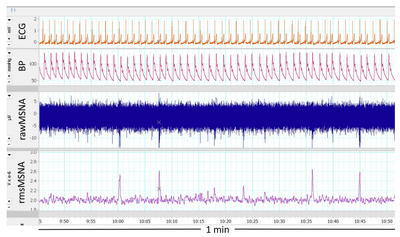
Results



Resting

HR: 47 b/min

MAP: 74 mmHg

MSNA: 7 bursts/min

One minute of data from a young female during a 10-minute resting phase.HR: heart rate; MAP: mean arterial pressure; MSNA; muscle sympathetic nerve activity. Values represent a one-minute average.

Head Up Tilt Test

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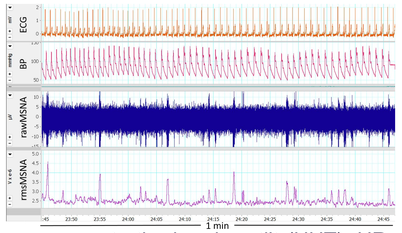

One minute of data from a young female in response to the head up tilt (HUT). HR: heart rate; MAP: mean arterial pressure; MSNA; muscle sympathetic nerve activity. Values represent a one-minute average.

Discussion

- Preliminary data demonstrates an increase in HR, MAP, and MSNA with the head up tilt test.
- We expect a decrease in sympathetic function with VNS.
- Decrease in these measures will demonstrate that modulation of the vagus nerve afferents can influence the autonomic nervous system and potentially lead to therapeutic treatments.

Conclusions

The REVEAL study systematically investigates the autonomic, cardiovascular, immune, and metabolic systems using various Vagus Nerve Stimulation (VNS) parameters in participants with FDA-approved indications: depression and epilepsy.

Future Investigations

The findings inform the development of more effective VNS therapies and thus advancing treatment options for these conditions. This research holds promise for refining VNS interventions and expanding their applications with potential implications for a wide range of medical domains such as kidney disease, gastrointestinal disorders, cardiovascular diseases, and more.

Literature Cited

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Acknowledgements

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Research Evaluating Vagal Excitation and Anatomical Linkages (REVEAL) on the effect of Vagal Nerve Stimulation

Insia Kizilbash[1], John Osborn[2], Miguel Anselmo[3], Emma Lee[1], Manda L. Keller-Ross[1,3]

[1]Division of Physical Therapy, [2]Division of Surgery, [3]Division of Rehabilitation Science, University of Minnesota

Minnesota's Louis Stokes Alliance for Minority Participation – MnDRIVE

University of Minnesota – Driven to Discover℠

Introduction

- Vagus Nerve is the primary nerve involved in parasympathetic function.
- Vagus Nerve Stimulation (VNS) is approved to treat epilepsy (1997) and depression (2005), however, its influence on physiological systems is unknown.[1]
- The autonomic nervous system (ANS) maintains homeostatic control in the body and regulates systems like thermoregulatory, cardiovascular, and respiratory systems.[2,3]
- ANS consists of sympathetic and parasympathetic activity that work interdependently.
- Sympathetic nervous system (SNS) activity increases with disease; high SNS can lead to end organ damage.
- How vagus nerve stimulation influences SNS activity is unclear, but could lead to novel and innovative therapies.

Purpose & Hypothesis

Purpose: Use a variation of VNS stimulation parameters to determine acute and chronic effects of VNS on sympathetic function.

Hypothesis: We hypothesize that VNS stimulation will reduce sympathetic activity at rest and during ANS perturbations.

Methods

General Overview of REVEAL Clinical Trial

- The REVEAL study is a multi-site, single-blinded, crossover study.
- The target sample size is 144 participants of 18 years of age or older.
- Six parameters of acute and chronic VNS are received by participants in a randomized order.
- Measuring various parameters throughout help us study in particular the cardiac and autonomic systems in relation to VNS.

Specific to the Aims of this study

- Participants will attend two visits with continuous monitoring of BP, sympathetic activity, and HR using parameters of VNS during rest, exercise, and head up tilt.

Figure 1. Microneurography procedure to measure muscle sympathetic nerve activity.

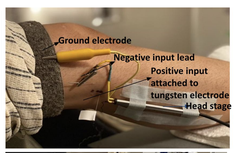
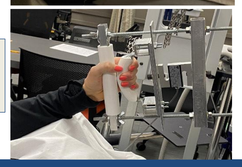


Figure 2. Isometric Handgrip Exercise. Used to perform upper extremity fatiguing exercises with PECO.



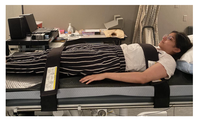
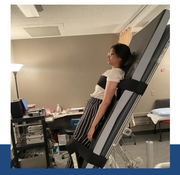


Figure 3. Head Up tilt. A) No incline B) 60° incline



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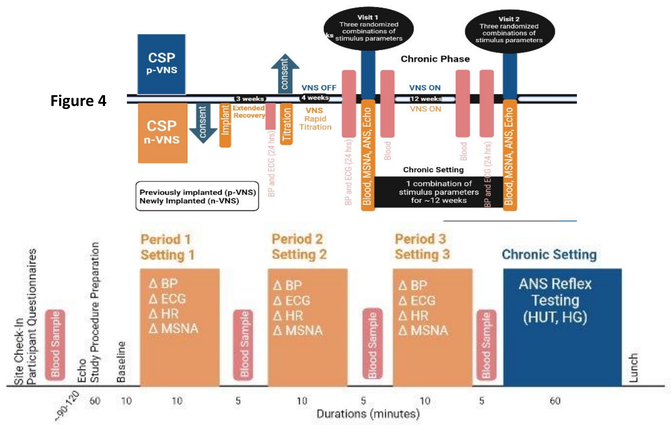


Figure 4

Figure 5a

Visit 1 Timeline

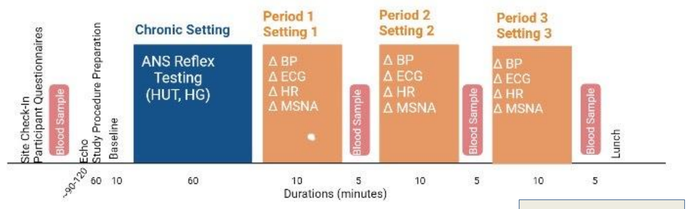
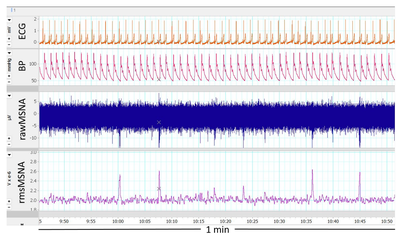


Figure 5b

Visit 2 Timeline

Created in BioRender.com

Results



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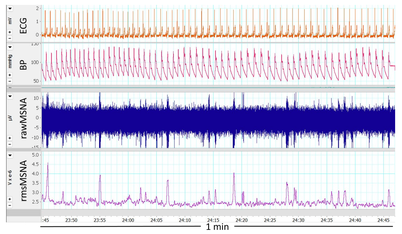

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The findings inform the development of more effective VNS therapies and thus advancing treatment options for these conditions. This research holds promise for refining VNS interventions and expanding their applications with potential implications for a wide range of medical domains such as kidney disease, gastrointestinal disorders, cardiovascular diseases, and more.

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Defining Immune Targets in response to Military Burn Pit Exposure in the Lung-Brain Axis

Jesus Romero Lopez [1,2], Sary Rios Diaz [1,6], Hunter Lo [1,7], Valerie Nguyen[1,12], Simon W. So [1,3], Jedidah J. Taylor[8], Joshua P. Nixon [1,4,5], Janeen H. Trembley[1,9], Julie M. Tomáška[10], Timothy R. Nurkiewicz[11] Tammy A. Butterick [1,3,4]

1. Minneapolis Veterans Affairs Health Care System, Minneapolis, MN, 2. Department of Biology , University of Minnesota Twin Cities, Minneapolis, MN , 3. Department of Neuroscience , University of Minnesota Twin Cities, Minneapolis, MN, 4. Department of Food Science and Nutrition, University of Minnesota Twin Cities, St. Paul, MN, 5. Department of Surgery, University of Minnesota Medical School, Minneapolis, MN, 6.Department of Biology, St. Olaf College, Northfield, MN, 7. Department of Biology, Saint John's University, Collegeville, MN, 8.School of Public Health and Tropical Medicine, Tulane University, New Orleans, LA, 9.Department of Laboratory Medicine and Pathology, University of Minnesota, Minneapolis, MN, 10. Burn Pits 360, Robstown, TX, 11. University of West Virginia, Medical Center for Inhalation Toxicology, Morgantown, WV, 12. Department of Psychology, University of Minnesota Twin Cities, Minneapolis, MN.

Introduction: Millions of troops during the Afghanistan and Iraq wars were exposed to burn pits. Large areas where materials and garbage were disposed of by burning them (1,3,4,5,13).After burn pit exposures veterans reported multiple symptoms such as respiratory problems, mood and cognition changes, fatigue, and other bodily pain or changes (1,5,13) Studies and analysis of inhalation of toxic fumes cause physical damage (5) and inflammatory responses that spread to other organ systems (13). Rat models allowed for whole body inhalation exposure using carbon black (CB) particulate form of elemental carbon manufactured by the gas-phase pyrolysis and partial combustion of hydrocarbons (13). Further studies included naphthalene (NA) a polycyclic hydrocarbon that is a biohazard and suspected carcinogen (9) that mimics aromatic hydrocarbons from burn pits. Showing that CB/NA inhalation illicit immune responses in the brain and lung.

Hypothesis:

We hypothesize that NF-*k*B, a transcription factor involved in various cellular process and inflammatory responses (8) plays a role in upregulating other inflammatory responses and drives inflammation after CB/NA inhalation. Thus, NF-*k*B will be present in the nucleus of brain and lung cells.

Objectives:

1. Determine the presence of cytokine in brain and lung tissue after CB/NA inhalation.
2. Determine the presence of NF-*k*B in the nucleus of brain and lung cells of rodents exposed to CB and NA.

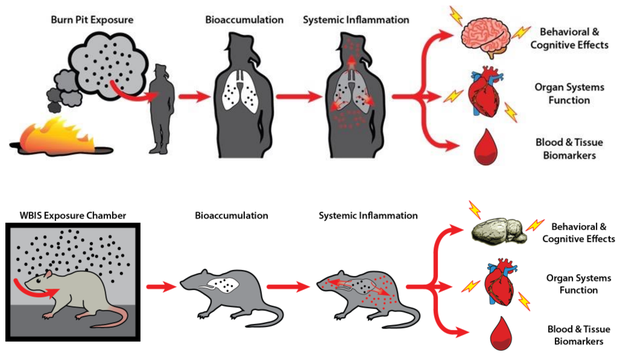

Figure 1. Conceptual model used to mimic burn pit exposure of humans by exposing rodents to CB and NA. Figure modified from Taylor et al.(12).

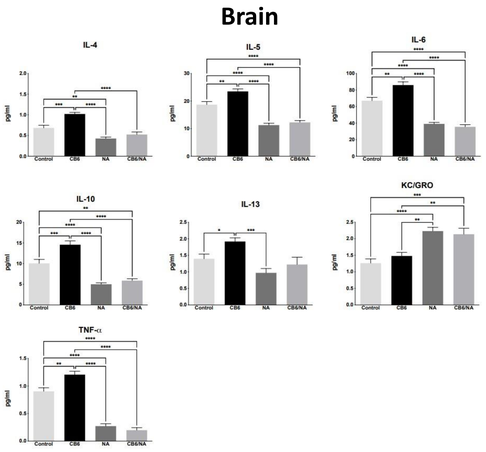

Figure 2.Brain proinflammatory biomarker analysis in rats after CB and NA 6 hr exposure (6 mg/m3). Significance is denoted as follows:* p<0.05,** p<0.005, *** p<0.0005, ****p<0.0001.

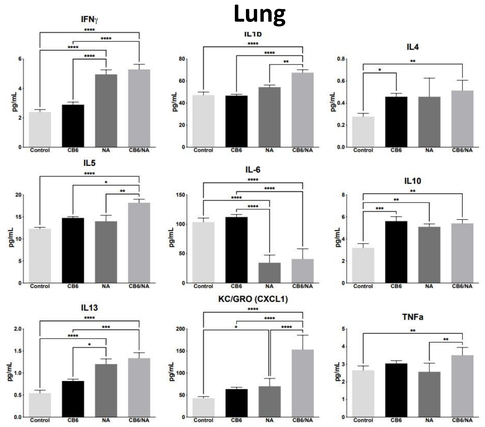

Figure 3. Lung proinflammatory biomarker analysis in rats after CB and NA 6 hr exposure (6 mg/m3). Significance is denoted as follows:* p<0.05,** p<0.005, *** p<0.0005, ****p<0.0001.

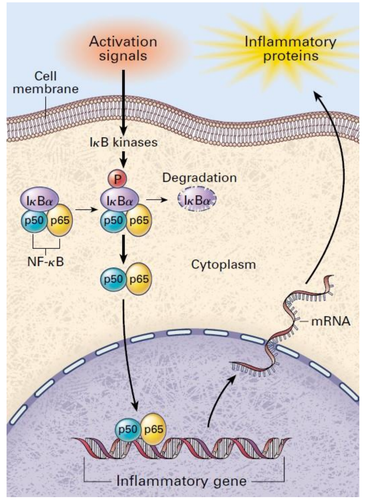

Figure 4. Diagram of NF-*k*B activation pathway. Image modified from Barnes and Karin(2).

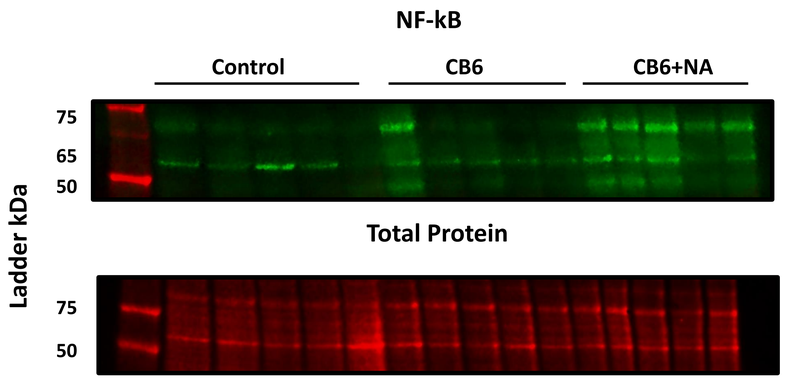

Figure 5. Total protein and NF-kB Western blots.

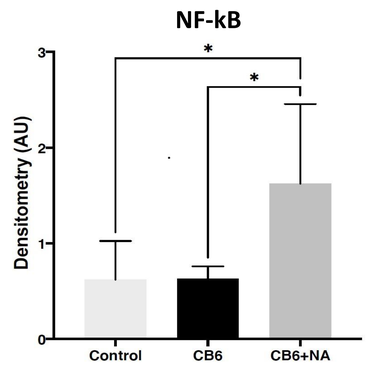

Figure 6. Densitometry normalized to total protein. Significance is denoted as:* p<0.05

Scan for Additional Resources

Conclusion:

- Rodent models provide a surrogate for burn pit exposure.
- Proinflammatory biomarkers were found after CB/NA exposure in rat studies
- Western blot results NF-kB show a significant increase in CB/NA than c control and CB6 samples alone.

Methods:

Whole body inhalation exposure, tissue collection and processing, and immunoassays were performed as previously described in Trembley et al(13) . Butterick/Nixon lab protocols were used for Western blots and as previously described in So et al(11). One Way ANOVA followed by Tukey's comparison tests were used for Western blot statistics.

Discussion:

- Future projects and studies involve bulk RNA sequencing of tissue samples and exposing microglial cells to heavy metals.
- Lack of treatment for burn pit exposure. Therapies to alleviate inflammation, such as sauna use, need further exploration. As studies have shown to have anti-inflammatory and positive cognitive benefits (6,7,10,14).

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From City Centers to Rural Areas : A Study of Roadside Plant Species Richness and Diversity Across an Urban Gradient

Layevska Jiménez-Hernández[1], Luis Santiago-Rosario[2], Emilie Snell-Rood[2]
[1] Carleton College, Northfield and [2] Department of Ecology, Evolution and Behavior, University of Minnesota, St. Paul

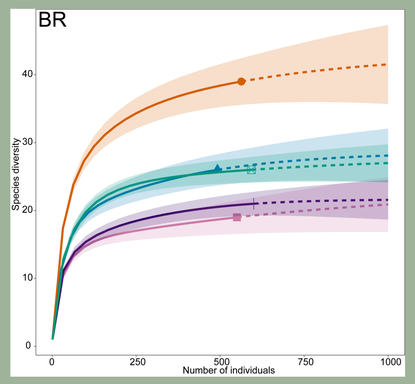
Background

Urbanization increases the **competition** for space and resources, **threatens** biodiversity and advances the **disruption** of natural habitats.

The urban gradient provides a unique opportunity to study how plant communities respond to varying levels of urbanization, ranging from the urban core to suburban areas, and eventually to more undisturbed rural environments.

How do different degrees of urbanization intensity influence roadside plant communities?

Results



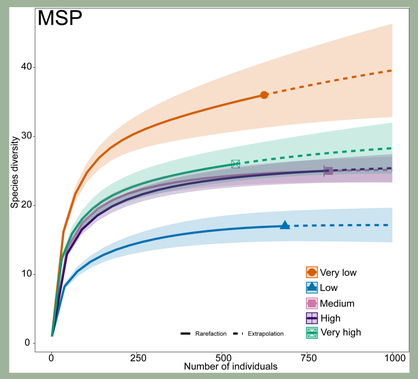


Figure 2a and 2b Rarefaction curve representation to depict how the number of different species observed increases with the increasing number of individuals collected in the BR and MSP sites.

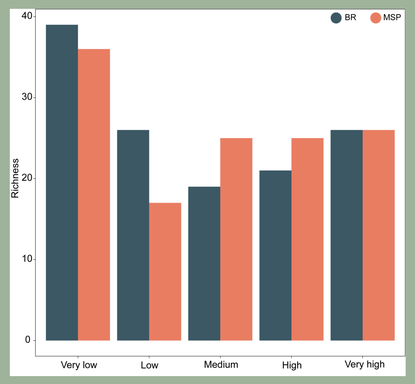


Figure 3a. Bar graph showing species richness for each community and city.

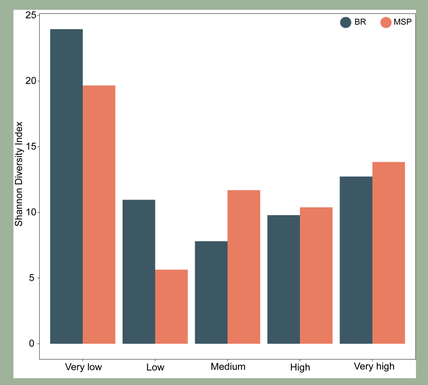


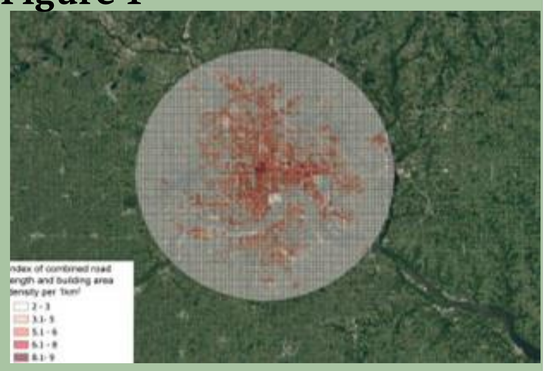
Figure 3b. Bar graph showing plant diversity, using Shannon index for each community and city.

Methods

Site Selection

For Baton Rouge, La, and the Minneapolis-St. Paul Metropolitan area, we delimited a 40-mile radius from city center. The radius was segmented into 1km²-grids and for each grid the building area and the sum of road lengths were calculated and made into an index. Then, the indexed for each were summed to create an urbanization degree index divided into 5 categories. (Figure 1).

Figure 1



Plant community sampling protocol:

- A 0.5m x 0.5m quadrant was used to isolate three sets of plots per site
- iNaturalist was used to identify species
- For each plot, each species and its abundance was quantified.
- A rarefaction curve was generated for each community

Discussion

Initial expectations included a steady decline in species richness and diversity as urban density increased.

- Lower urbanization degree in suburban areas may influence the presence of high species diversity.
- Decrease in species diversity due to consistent maintenance in suburban gardens and roadsides.
- Invasive species may be contributing to increase species richness and diversity

Well-adapted species thrive, promoting survival and reproduction.

Both rarefaction curves for the BR and MSP data results show plant species diversity was at its highest at very low urban density areas.

Notably, at low-high level urban density, MSP data for species diversity was lower than observed in BR data.

Further Research

- Foster understanding and conservation efforts in biodiversity in urban environments for sustainable urban development.
- Exploration on how plants have become adapted to gain resilience towards urbanization.
- How and which Urban drivers impact species richness
- The value of Shannon diversity and specie richness when looking at ecological health
- Indicators of ecological instabilities

Acknowledgments

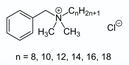
I'd like to thank Luis Santiago-Rosario for his instrumental guidance and expertise in shaping the direction of this study as well as the unwavering support and collaborative atmosphere of the Snell-Rood Lab.

How the Compounds Potassium Permanganate and Potassium Ferrate Eliminate Water Pollution

By: Maeva Tchouaffe, Dr. William A. Arnold, & Anna Mahony

Introduction

- The goal of this project is to determine how efficient potassium permanganate and potassium ferrate are in decreasing C12BAC concentrations from wastewater treatment plants, and in turn, reducing water pollution overall.

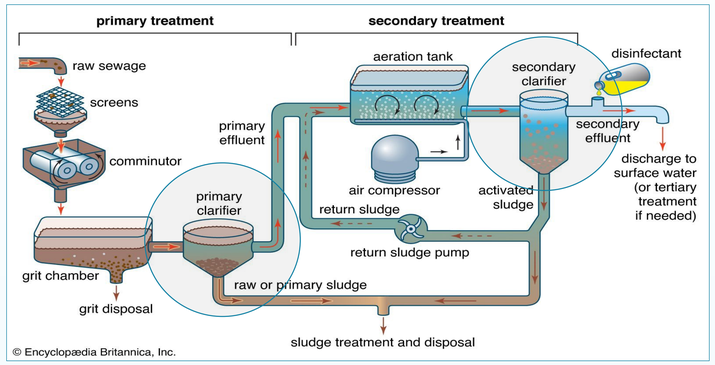


n = 8, 10, 12, 14, 16, 18

- C12BAC (a benzalkonium chloride structure with a carbon chain of 12) is a type of QAC, which stands for quaternary ammonium compound. QACs, also referred to as quats, always have two components: a central nitrogen atom, and 4 clusters of atoms tied to the central nitrogen atom. They are commonly found in everyday disinfectant products, and in wastewater treatment plants effluents, all over the world.
- A comprehensive screening experiment of wastewater effluents and lake sediments, ran in 2018, concluded that the level of QACs, found in both effluent water and sediments, could pose an issue over time, specifically, antibiotic resistance and water pollution
  - The results revealed high concentrations of QACs in both the effluent and sediment samples collected, supporting the hypothesis effluents from WWTPs are depositing QACs into natural waters.
- A review lead by Yufen Wang concluded that potassium permanganate could be used as a QAC decontaminant, through oxidation, due to its many advantages being highly efficient, affordable and nontoxic.
- Similarly, a study led by Yunho Lee concluded that potassium ferrate could also be used as a QAC decontaminant, through oxidative elimination as well.
  - Example of oxidation process:
    MnOMnO4- + QAC --> MnO2 + oxidized QAC

Scientific Question & Hypothesis

Can potassium permanganate and potassium ferrate be incorporated in the primary and secondary clarifiers of wastewater treatment plants process, in order to reduce water pollution caused by QACs?

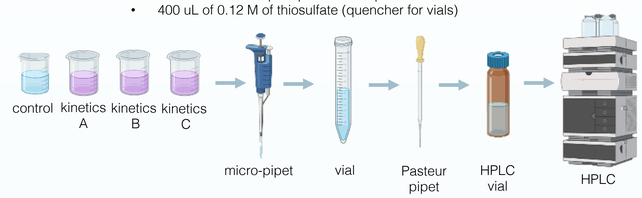


Hypothesis: The concentration of C12BAC will decrease in both the permanganate kinetic experiments and ferrate absorbance experiments, due to the oxidation of both oxidants.

Method

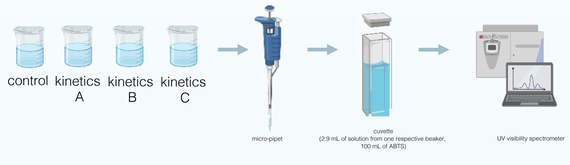
Permanganate Experiment

- 1 mL of 50 uM of C12BAC added to each beaker
- 1 mL of 50 uM of potassium permanganate added to kinetic beakers
- 98 mL of phosphate buffer pH 7
- 400 uL of 0.12 M of thiosulfate (quencher for vials)



Ferrate Experiment

- 1 mL of 25 mM of C12BAC added to each beaker
- .5 mL of 5 mM of potassium ferrate added to kinetic beakers (1 mL)
- 98.5 mL of phosphate buffer
- 100 uL of 15 mM of ABTS (quencher for vials)



Results

1.

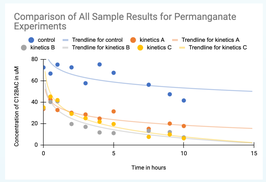


Figure 1 shows the results of the permanganate experiments and includes the graphs for the control and kinetic A, B, and C experiments. A natural log trend line is depicted for all four data sets, from hour 0 to hour 10. The data was collected and documented in Google Sheets, and the graph was made using Google Sheets as well.

2.

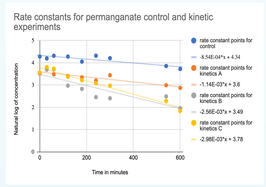


Figure 2 depicts the rate constants of the permanganate experiments and includes the k values (the slope value in each equation) for the control and kinetic A, B, and C experiments. Time is measured in minutes, but still ends at 10 hours. The data was collected and documented in Google Sheets, and the graph was made using Google Sheets as well.

3.

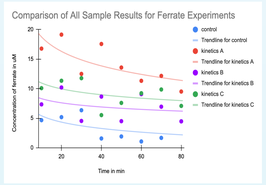


Figure 3 shows the results of the ferrate experiments and includes the graphs for the control and kinetic A, B, and C experiments. A natural log trend line is depicted for all four data sets, from minute 0 to 80 minutes. The data was collected and documented in Google Sheets, and the graph was made using Google Sheets as well.

4.

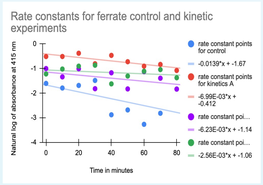


Figure 4 depicts the rate constants of the ferrate experiments and includes the k values (the slope value in each equation) for the control and kinetic A, B, and C experiments. Time is measured in minutes, and ends at 80 minutes, like in figure 3.The data was collected and documented in Google Sheets, and the graph was made using Google Sheets as well.

Conclusion

Overall, the results depict a downward trend, which supports our hypothesis. However, the data that makes up the results is inconsistent, meaning future experiments and research will need to be conducted, in order to achieve more accurate results.

For the permanganate experiments, although a downward trend is present, both the results for the control and the kinetic experiments, show peaks in the concentration of C12BAC, after its concentration had decreased prior. This is especially concerning for the control in the permanganate experiment, which should have a near constant concentration of C12BAC, considering that potassium permanganate was not present. Additionally, the control has concentrations that are two times higher than the kinetic experiments, which can be explained by an error during the experiment i.e., adding two times the amount of C12BAC required.

Similarly, for the ferrate experiments, a downward trend is also present for both the control and kinetic experiments, however, it is not a consistent downward trend, with peaks in the concentration levels of after a decrease in ferrate concentration. Unlike the permanganate experiments, the control for the ferrate experiments did not have any C12BAC and was simply potassium ferrate in pH 8 phosphate buffer. Potassium ferrate in pH 8 phosphate buffer has a short half, of no greater than an hour and thirty minutes. Therefore, the control samples showing a decrease in ferrate concentrations was expected, however, the peaks were not.

Limitations

1. Lack of other experiments pertaining to the use of both compounds reducing C12BAC
2. Sorption element of QACs → potential sorption to glass stir rods, beakers, vials, pipet tips and plastic pipet tips
3. Potassium ferrate's rapid half life → decrease in concentration is inevitable, difficult to measure if it's due to C12BAC

Future Experiments

For the permanganate experiments, a new kinetic experiment has been completed and will be analyzed shortly. This experiment collected data from a control in a glass beaker (with the accurate amount of C12BAC), as well as two other samples where the C12BAC, potassium permanganate, and phosphate buffer solutions were held in polycarbonate bottles. Polycarbonate bottles were substituted for the glass beakers in the new experiment because C12BAC should not sorb to those bottles, therefore, the samples could return more accurate results on C12BAC concentrations. Two polycarbonate bottle were used for this experiment: one that held a control solution, identical to the one in the glass beaker, and the other that held a kinetic solution, identical to the ones in the glass beakers, from previous experiments.

For the ferrate experiments, discussion about future experiments is yet to be held, but double checking that C12BAC does not sorb to the material that make up the cuvettes, would be a great start. Additionally, finding a buffer that would slow down the half life of ferrate, would be hugely beneficial in measuring accurate future ferrate concentrations.

Acknowledgements & References

Project supported by the Louis Stokes North Star STEM Alliance, National Science Foundation award number CON000000064472, and the University of Minnesota's MnDRIVE (Minnesota's Discovery, Research, and InnoVation Economy) initiative.
University of Minnesota
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Minnesota's First Cash Cover Crop
Can Fungi Produce Better Food?

Marco Moriarty, Xiao Sun, Kristin Boardman, and Bo Hu

Department of Biosystems and Bioproducts Engineering, University of Minnesota – Twin Cities

Abstract

Farming has negative effects on the environment. During farming, after harvest, a lot of nutrients are left in the field, which wash away during the Spring. In drainage water, these nutrients act as a fertilizer to algae, which reduces biodiversity. A common solution is cover crops, which hold in the nutrients and protect the fields. **However, these cover crops are not usually cultivated because they are more work and don't have an economic value.**

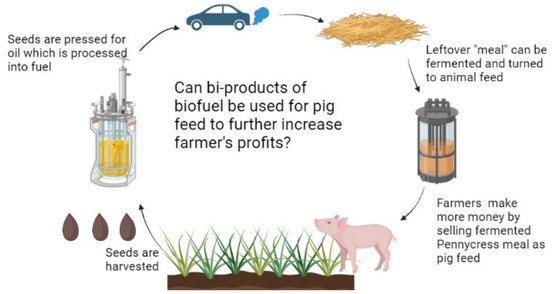
There needs to be a crop that can (1) be grown in late fall, (2) holds the nutrients like a regular cover crop, and (3) can provide economic value.

Pennycress is the perfect candidate as their seeds have up to 33% oil production and can be used for biofuels.

Key Terms:
- Fermentation: Fungi and microbes use a substrate (Pennycress) as a food source. They break down either sugar or protein as a fuel source and release chemical biproducts.
- Feed: Food for animals.
- Biodiversity: The variability of lifeforms in an ecosystem.

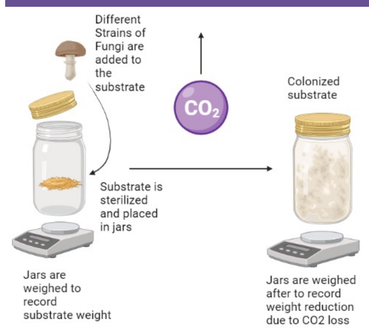
Introduction

Pennycress seeds have oil which can be converted into biofuel. However, after the oil has been pressed, there are still nutrients that may have economic value. The point of our experiment is to find out if the leftover "waste" can be converted into food for animals.



This material is high in fiber and protein. The fiber is hard for the animals to break down and the protein quality is low. **It may be possible to use fungi to break down fiber, and enrich protein content, converting non-essential amino acids into essential amino acids.**

Methodology



Total Weight Loss –How much biomass is lost during fermentation?(Due to CO2)

Substrate is autoclaved in a jar to prevent contamination, then is fermented.
Jar and substrate are weighed before and after to record weight loss due to fermentation.

Figure A. Process for total weight loss analysis.

Methodology

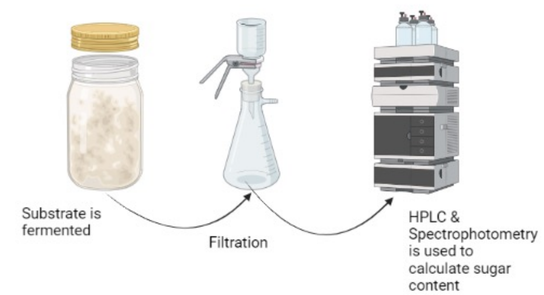


Figure A. Process of fermentation and sugar content analysis

Reducing Sugar: Can fungi reduce sugar content? What can sugar reduction tell us about growth rate?
Substrate is fermented then filtered.
Filtrate and liquid are both tested for sugar content. Methods for sugar testing are spectrophotometry and HPLC.

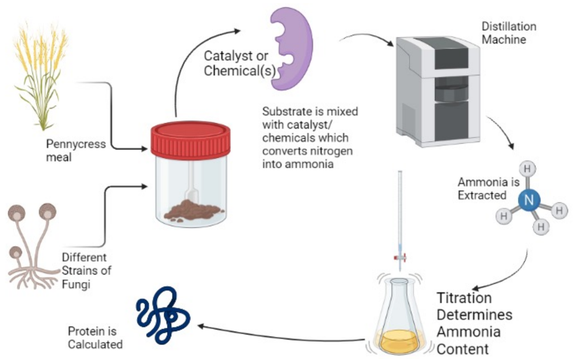


Figure B. Process of fermentation, digestion, & protein analysis

Total Protein: Can fungi turn non-essential proteins into essential amino acids?
TKN:
Ferment Pennycress meal
Digest the fermented meal to convert any organic Nitrogen into Ammonia
Distill out the ammonia
Use titration to get a measurement of ammonia nitrogen to calculate total nitrogen.
Multiply Nitrogen by standard to get total protein
TAN:
Distill fermented sample to get ammonia existing in the sample
Use titration to get a measurement of ammonium nitrogen
(TKN – TAN) * Factor (6.25) = Total Protein

Results

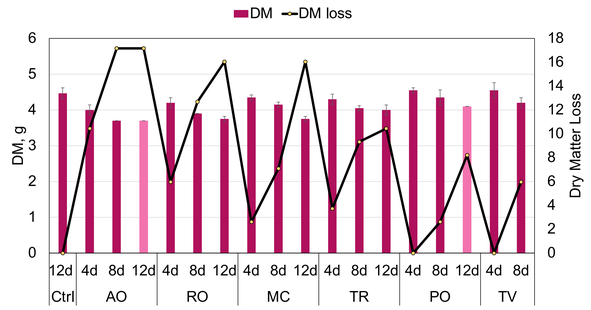


Figure 1: Total Mass loss over time.

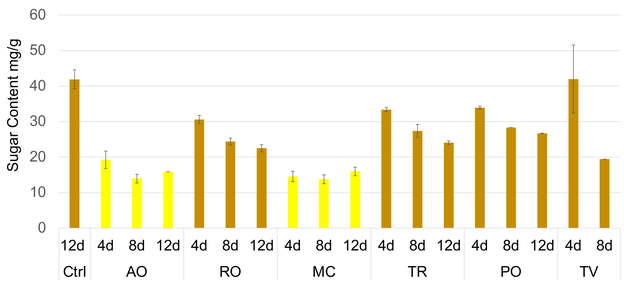


Figure 2. Reducing Sugar during fermentation.

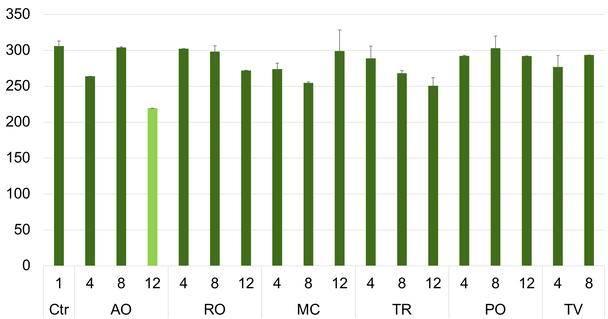


Figure 3. Total Crude Protein

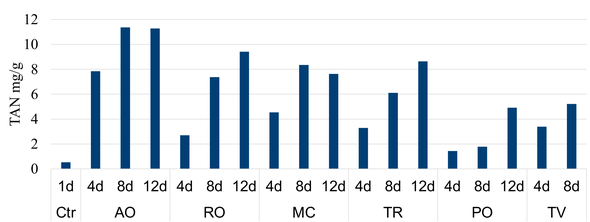


Figure 4. Total Ammonium Nitrogen

Discussion

During fermentation, carbon is released. Ideally, less carbon loss is better as it produces more feed. All strains showed about an equal amount of dry matter loss, but AO, RO, and MC showed the most amount of reduction. Longer fermentation times resulted in more dry matter loss.

Fungal strains AO and MC had the highest amount of sugar reduction.

Sugar can be produced by breakdown of complex carbohydrates, which may have resulted in certain strains producing more sugar during later fermentation times. This would explain why our measurements show sugar going up over time. However, these changes in sugar are non-significant.

For protein, AO had the highest amount of protein reduction. Too much protein reduction results in lots of ammonium being produced, which is not good for animals. We want protein reduction, but not too much.

.

Conclusion

TR showed moderate weight loss with high sugar reduction and moderate protein reduction. We want protein to be reduced, but not all the way, because that would increase the ammonium content. TR seems to be the most promising for our objectives.

Further tests will need to be done to figure out how to ferment Pennycress at large scale, while still achieving optimal nutrient breakdown rates.

Further experiments would test the optimal number of days for fermentation, recording data every two days instead of four.

In future tests, extra nitrogen would be added to see how that would affect total protein content.

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Acknowledgements

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Preliminary study: The Effect of Rhyme context on Word Recall in a complex span divided attention task

Marta Bacon, Carleton College

MnDRIVE Mentor (2023): Dr. Jayanthi Sasisekaran, Associate Professor, Department of Speech-Language-Hearing Sciences
University of Minnesota – Twin Cities

Significance

- **Phonological Similarity Effect (PSE):** The phonological similarity effect is the inhibitory effect of similar sounding words on word recall tasks.
- This study investigated if a rhyming context inhibited (PSE) word recall in a version of the complex span reading task involving (1) word recall and (2) sentence judgement.
- Previous research suggested PSE is influenced by several task factors. This study investigated individual differences in complex span working memory and the processes/mechanisms mediating PSE.

Hypothesis: Rhyming context inhibits word recall in both individuals with higher vs. lower working memory (HWM vs. LWM) and potential group differences are due to differences in phonological vs. attentional mechanisms supporting word recall in the complex span task.

- Task 1: We found differences among participants categorized into HWM vs. LWM (based on their complex span word recall scores). Those in the HWM group were able to use articulatory rehearsal more effectively to overcome PSE and accurately recall words. In contrast, the LWM group showed larger PSE.
- Task 2: We also investigated if PSE influenced performance in a sentence task in which they had to respond if a sentence made meaningful sense or not.

Introduction

- Previous research that reported that phonological similarity facilitates **word recall** or **working memory required** participants to complete a simple span task (Figure 1).
- If they used a complex-span model, the rhyme vs. nonrhyme context cue was provided by the words in the recall list. In this study the context cue was provided by the last word of each sentence that either rhymed or not with the word preceding it.
- Decision-making and judgment requires an interaction between the hippocampus and other regions of the brain supporting working-memory processes.
- **The mechanisms of this action and the influence of other brain regions are not well understood.** The altered version of the complex span task allows us to investigate the influence of PSE in this process.

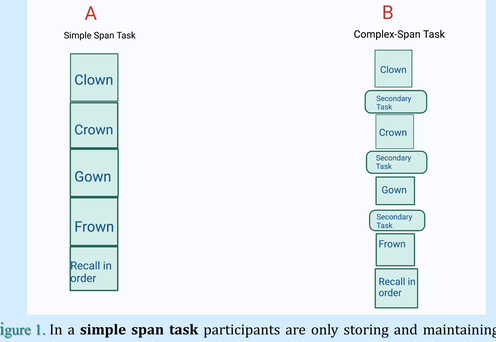


Figure 1. In a **simple span task** participants are only storing and maintaining items in their memory; they are asked to recall a series of stimuli directly after the last item is presented (A). **Complex-span tasks** are more representative of working memory as they are designed to involve a second task in between storing and maintaining items. The time interval introduced by the complex-span task requires not only storing and maintaining items in memory but also to divide attention with another task (B).

Methodology

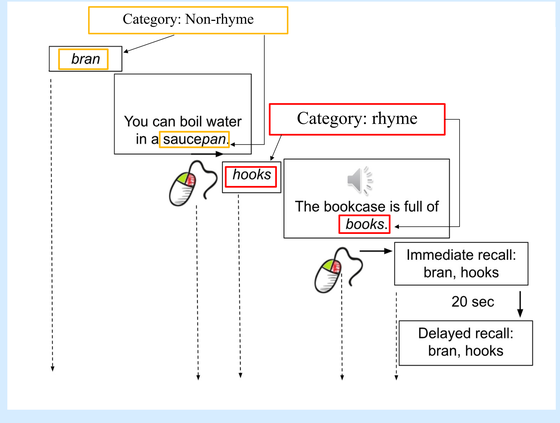
Participants

- 10 young adults without a current history of language reading or speaking difficulties.
- Participants were categorized into Higher (HWM) vs. Lower Working Memory (LWM) groups based on group mean in word recall from the experimental task.
- Participants in both groups were similar in age, expressive vocabulary, forward digit span and sentence recall.
- The groups performed differently in the backward-digit span task, although this did not correlate with performance in the experimental task.

Complex-Span Task

Levels 2-5

Task 1	Task 2
Word recall	Sentence judgment



Dependent Measures

I. Response time at each level and category from the sentence task.

II. Accuracy at each level and category from the word recall task.

Results

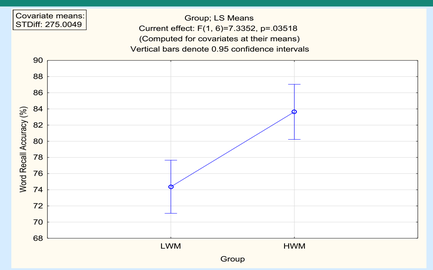


Figure 2: Were the groups different in Complex Span Word Recall?

There was a significant difference in performance on the word recall task between the HWM and LWM groups. The HWM group (group 2) had significantly higher word recall scores than the LWM group (group 1).

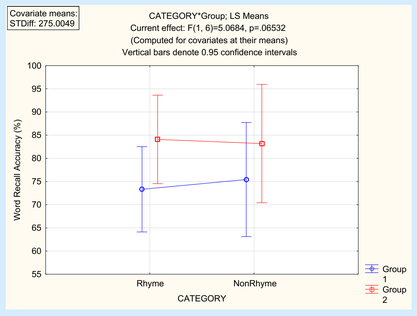


Figure 3: Is the difference category based?

There is category related influence on group performance in the word recall task. The HWM group had significantly higher word recall scores in the rhyme category (Category 1) than the LWM group (p=0.02). The descriptive group differences observed for the non-rhyme category were not significant (p=0.07).

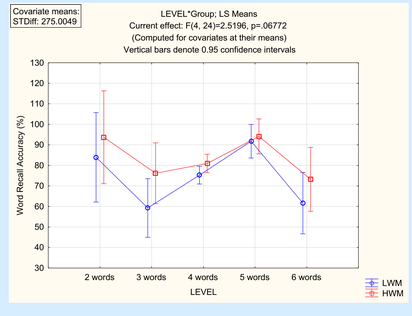


Figure 4: Is the difference level based?

There is level related influence on group performance in the word recall task. The LWM group showed larger differences in word recall scores at both the lower and higher levels of the task.

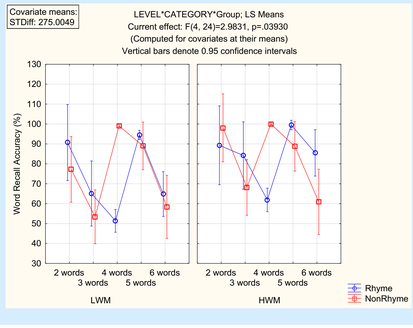


Figure 5: Were there effects of both category and level on group performances?

In the rhyme context category (category 1), the HWM group (group 2) performed better at the lower and higher levels. The HWM also showed better performance at the lower levels of the non-rhyme context category.

Results cont.

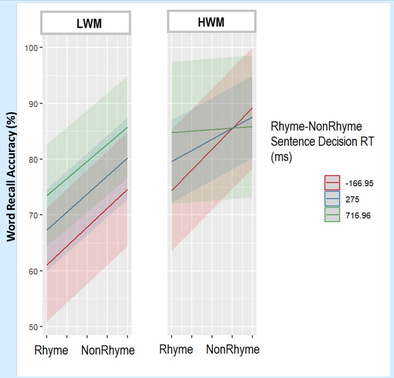


Figure 6: Is there a correlation between performance on the Word Recall task and the sentence task between groups?

The HWM group recalled words in the non-rhyme category with near comparable accuracy to the LWM group without being influenced by performance in the sentence judgement task All other comparisons showed speed-accuracy trade-off between sentence judgment and word recall, that was most evident in the rhyme category.

Conclusion

The observed group differences are attributable to executive function mechanisms.

- Preliminary differences suggest that the HWM group was more effective in overcoming the PSE effect in the rhyme condition.
- The findings also suggested that the HWM group utilized the attentional mechanisms supporting maintenance for word recall effectively in the nonrhyme condition.

The category and level based effects on group performance are near significant. Hence, replication of the findings in a larger sample is required.

Acknowledgements

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Social determinants of health effecting STEC infection among rural populations in Minnesota

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INTRODUCTION

- **Shiga toxin-producing *Escherichia coli* (STEC) are virulent foodborne and environmental bacteria[1] that pose a significant risk to populations under five years of age due to the possibility of developing hemolytic uremic syndrome (HUS) as a result of infection.[2]**
- **Rurality has been previously known as important factor in STEC infection rates.**
- **This study aims to evaluate what socioeconomic factors within rural communities are linked to the rate of cases in each exposure variable.**

METHODOLOGY

American Community Survey (ACS) data used from the US census website are mapped using ZCTAs (Zip Code Tabulation Area). A ZCTA is included in the study if it provides zip codes labeled as "neither" indicating rurality.

STEC infection rate was computed as the number of cases in the ZCTA divided by the ZCTA's population per 100,000 people from 2010 to 2019.

↓

Exposure variables were selected from 4 surveys on the ACS US census website. The summaries for each exposure variable will be stratified.

↓

A scatter plot was made for each exposure variable including the correlation. The variables were divided into quartiles summarized with frequency tables, and ANOVA test used to determine statistically significant differences between quartiles.

RESULTS

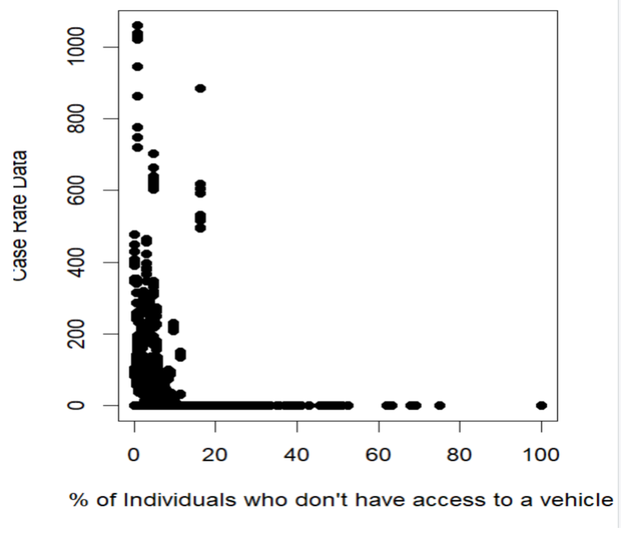

Figure A. This scatterplot depicts the connection between one exposure variable (the percentage of individuals who don't have access to a vehicle) and the case rate data that was previously calculated. The correlation coefficient is -0.057, indicating minimal negative correlation.

DESCRIPTIVES

Descriptives

	Under_5_quartile	case_rate_data
N	1	2852
	2	2852
	3	2852
	4	2852
Mean	1	10.54601
	2	17.64992
	3	14.08433
	4	16.01919
Standard deviation	1	37.11811
	2	69.76310
	3	44.84329
	4	50.52035

Figure B. This table shows the mean and standard deviation of the rate of STEC infections in rural zip codes within each quartile of the population under 5 years old. The mean case rate is lowest in zip codes with the fewest children under 5 years old. The ANOVA results indicated significant differences between quartiles, ($F = 9.857$, $p < 0.001$).

CONCLUSIONS

Understanding what sociodemographic factors within rural communities are linked to the rate of STEC cases in Minnesota is important for informing policy on how to prevent infection among groups who are at higher risk developing the infection. Factors such as financial inequality (including) access to adequate plumbing and working in certain industries may be linked to the increase in risk for developing STEC due to lack of access to cost effective, clean food products. Effective steps to continue looking into the accessibility of food-safe products could be a possible extension of this topic.

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ACKNOWLEDGEMENTS

Project supported by the Louis Stokes North Star STEM Alliance, National Science Foundation award number CON000000064472, and the University of Minnesota's MnDRIVE (Minnesota's Discovery, Research, and InnoVation Economy) initiative.

Identification and Assessment Toxicology of Heterotrophic Bacteria within a Cyanobacteria Colony

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Introduction

- **Cyanobacteria** are photosynthetic unicellular bacteria. They overgrow in freshwater producing algae bloom and toxins that are toxic to humans and other organisms making water **unsafe** to consume.
- **Heterotrophic bacteria** in water helps with natural degradation of cyanotoxins.
- It is unknown why cyanobacteria produce cyanotoxin, but it may be a resource for surrounding microorganisms, creating possible un-explored **mutualistic and antagonistic** relationships between cyanobacteria and the microbes around it.
- Using **identification**, **isolation** and **assessment** techniques for heterotrophic bacteria, we can see if they grow in the cyanobacteria's toxin (K and I spent media).
- This study will help us understand heterotrophic bacteria's mechanism of cyanotoxin **biodegradation**.

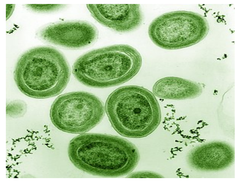

Figure 1. Image of algae bloom in stream (left). Microscopy image of cyanobacteria (right).

Goals

1. Identify heterotrophic bacteria in a cyanobacteria colony
2. Incubate the heterotrophic bacteria in the spent media
3. Assess the toxicology can help show potential of bacteria to biodegrade cyanotoxin in fresh water

Methodology

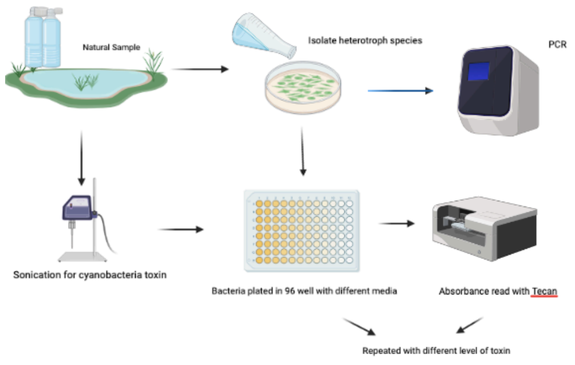

Figure 2. Outline of sampling, isolation of heterotrophic bacteria species, sonication of cyanobacteria to obtain toxin and plating species in a 96 wells with Tecan readings of absorbance.

Results

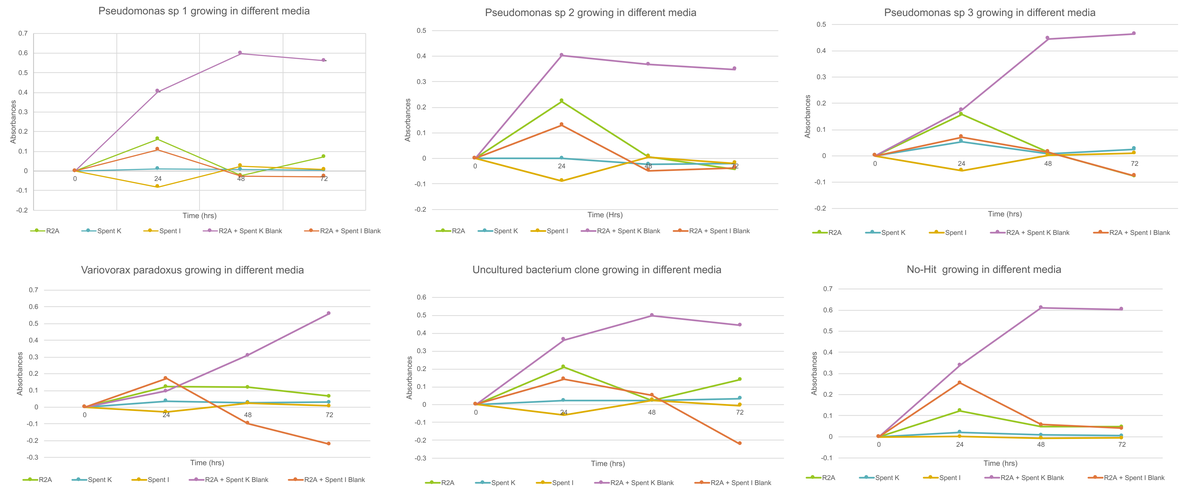

Figure 3. Each graph represents a different bacteria species grown in different media: R2A (green, nutrient rich media), Spent K (blue, cyanotoxin K), Spent I (orange, cyanotoxin I), R2A and Spent K (purple), R2A and Spent I (red) evaluated every 24hrs for 72hrs.

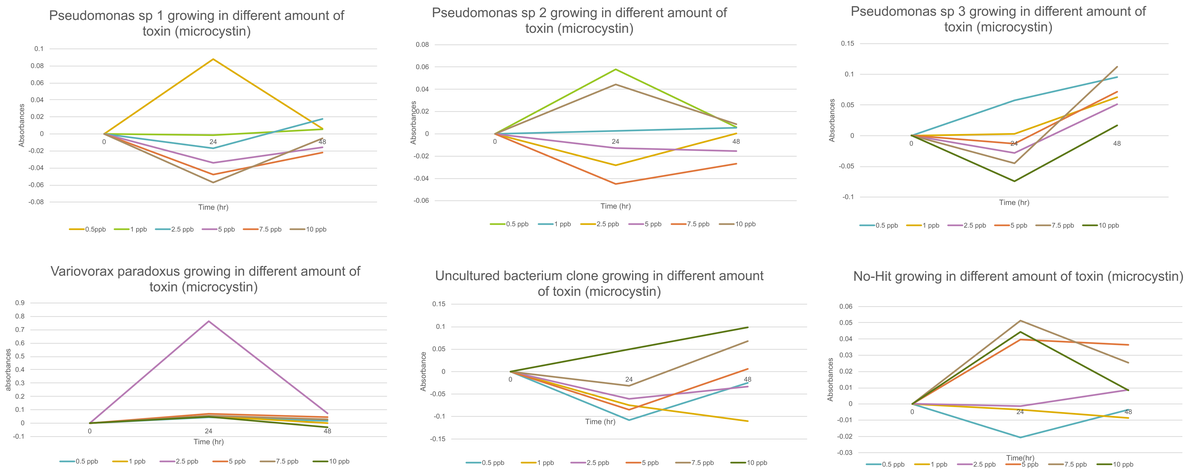

Figure 4. Each graph represents bacteria growth in different amount of microcystin toxin *ppb* (parts per billion) evaluated every 24hrs for 48hrs.

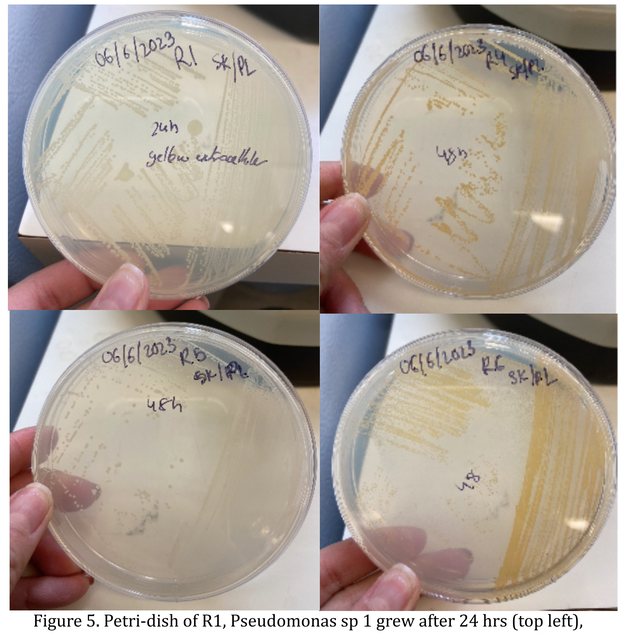

Figure 5. Petri-dish of R1, Pseudomonas sp 1 grew after 24 hrs (top left), petri-dish of R4, Variovorax paradoxus grew after 48 hrs (top right), petri-dish of R5, Uncultured bacterium clone grew after 48 hrs (bottom left) and petri-dish of R6 Pseudomonas sp 3 grew after 48 hrs

Conclusion

- Using isolation and the inability to do so shows that these heterotrophic bacteria are co-habitants with the cyanobacteria.
- We were able to identify Pseudomonas sp 1, Pseudomonas sp 2, Pseudomonas sp 3, Variovorax paradoxus and uncultured bacterium clone as five of the heterotrophic species using PCR.
- We found that all species growth the best in R2A and spent media K which contains the toxic from the original cyanobacteria.
- No-hit species, Uncultured bacterium clone, and Pseudomonas sp 2 shows that microcystin toxin does not inhibit their growth.

Next steps

- Repeat plating for Pseudomonas sp 1, 2, 3 and Variovarax paradoxus as we know they grow best in microcystin toxin based on the different medias.
- We will have to make controls for the ELISA experiment and use it to assess the biodegradation of cyanotoxin with all the bacteria that proves to grow in toxin.

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Impact of Military Burn Pit Toxin Exposures in the Lung-Brain Axis

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Introduction:

Millions of post-911 Veterans were exposed to toxic open air burn pits [2]. These burn pits used jet fuel to burn waste such as plastic, batteries, medical waste, amputated body parts, human waste, ammunition, and chemicals [5]. The burn pit emissions include hazardous particle matter which embed within the lungs and blood [9]. Research has associated a variety of health effects with burn pit exposures, such as chronic multi-symptom illnesses, respiratory problems, gross hematuria, lower urinary tract symptoms, and cancer due to chronic inflammation [6]. Inflammation is triggered by oxidative stress due to the burn pit environment. This activates nuclear factor-kB (NF-kB) and turns on the proinflammatory genes in the brain and lung [3].

Hypothesis:

Exposure of fine particle matter, carbon black and naphthalene, via a whole-body inhalation system will significantly increase an inflammatory response via the activation of NF-kB in a rat's brain and lungs tissue caused by oxidative stress similarly to the Veterans exposed to the burn pits [4].

Objectives:

1. Simulate burn pit exposure with a rat whole-body inhalation system (WBIS) model.
2. Determine if the activation of NF-kB in brain and lung tissue results in a proinflammatory response from oxidative stress.

Figure 1. Burn pit in US Base in Iraq [6].

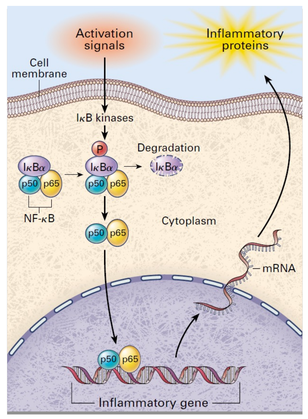

Figure2. NF-kB pathway diagram [1].

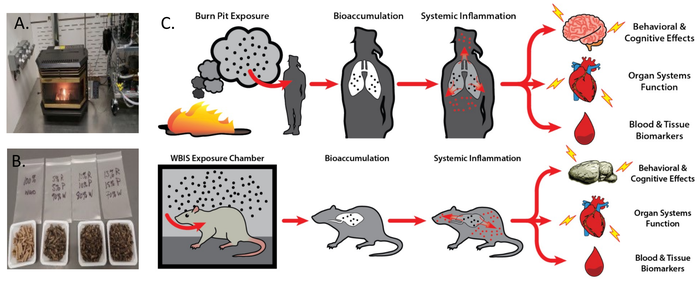

Figure 3. Model of burn pit exposure. (A) Circuit burn pit generator with JP8 jet fuel. (B) Mixed-waste material particle pellets. (C) Conceptual model [8].

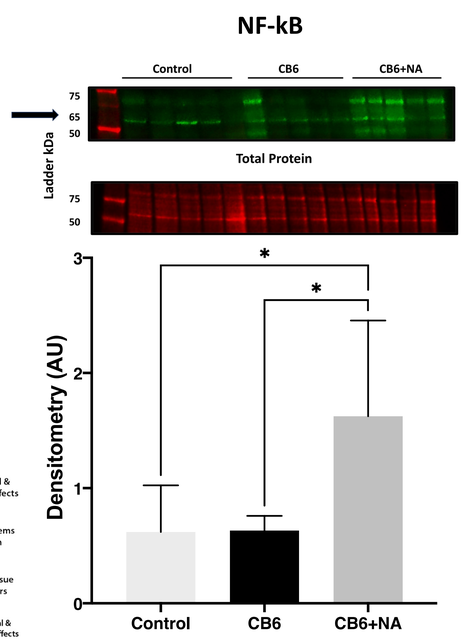

Figure 4. Total protein and NF-kB western blots from rat lung tissue exposed to carbon black (CB6) and naphthalene (NA).
Significance is denoted as: * p<0.05

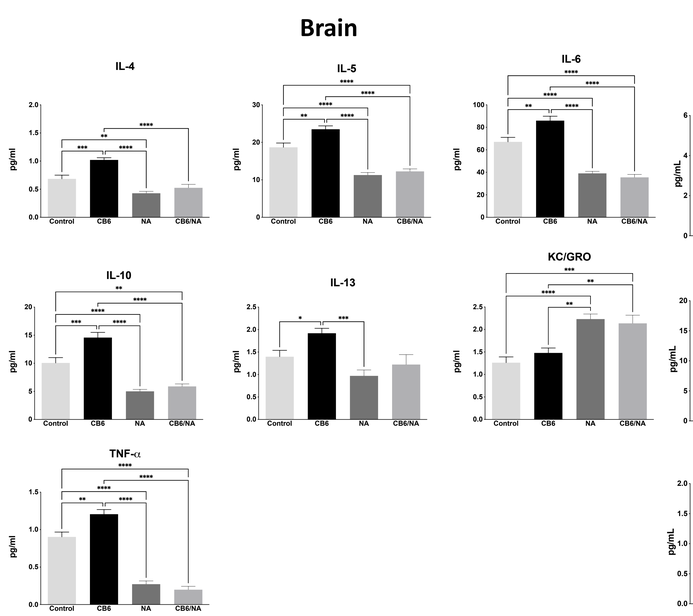

Figure 5. Brain proinflammatory biomarkers when exposed to carbon black (CB6) and naphthalene (NA) for 6 hours.
Significance is denoted as follows: * p<0.05, ** p<0.005, *** p<0.0005, **** p<0.0001

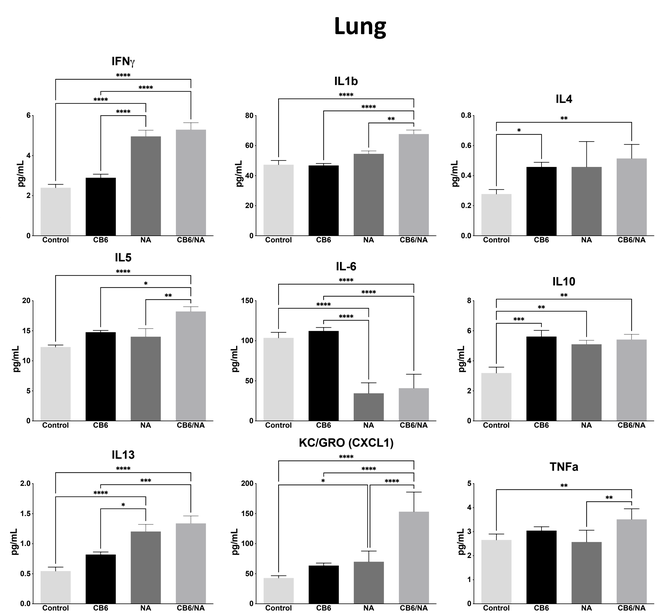

Figure 6. Lung proinflammatory biomarkers when exposed to carbon black (CB) and Naphthalene (NA) for 6 hours.
Significance is denoted as follows: * p<0.05, ** p<0.005, *** p<0.0005, **** p<0.0001

Discussion:

- When rat lung tissue were exposed to CB6 and NA, the western blots conveyed significantly more NF-kB in the CB6+NA compared to the control and CB6 group (Figure 4).
- There was a significant increase of the inflammatory biomarkers in the rat brain tissue when exposed to CB6 compared to control, NA, CB6+NA (Figure 5).
- Whereas in the rat lung tissue, there is a significant increase of the inflammatory biomarkers when exposed to NA and CB6+NA compared to control group and CB6 (Figure 6).

Conclusions:

- Results suggest NF-kB induces inflammation in rat lung and brain tissue when exposed to open air toxins.
- The significance of these results can pave a way to connect open air burn pit health effects on Veterans to communities in the U.S living in high atmospheric pollution.
- Future studies include more western blots on lung and brain tissue to measure immune cell activation and testing metformin drug on toxin exposed rats.

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Additional Information:

Methods:

Whole body inhalation exposure on rats, tissue preparation, biomarker screening: All methods and references available in additional information, references, entitled "Whole-body inhalation of nano-sized carbon black: a surrogate model of military burn pit exposure."
Western blots: All methods and references available in additional information, references, entitled "Early Life Obesity Increases Neuroinflammation, Amyloid Beta Deposition, and Cognitive Decline in a Mouse Model of Alzheimer's Disease."

Acknowledgements:

Project supported by the Louis Stokes North Star STEM Alliance, National Science Fund award number CON000000064472, and the University of Minnesota's MnDRIVE (Minnesota's Discovery, Research, and InnoVation Economy) initiative. This work was supported by the following sources: 2023 VA ORD BLRD Field Meeting Award, BX004146-05A1 (MAK, TAB, JHT, JMT, AG, AC, AMS)); Merit Award, VA ORD I01BX004655-04, (MAK); VA Lung Cancer Precision Oncology Program I50; VA ORD CU000163-01 (MAK), CDMRP DOD PR211133 (AMS); Supplemental Award Agreement MTRC 20211012.1605 (AMS); NIH R01 HL152385 (AMS); CDC NIOSH U01 OH012264-01 (AMS); VA Career Development Award IK2 CX001679 (JBL)

Developing a behavioral task that can assess if mice can differentiate more favorable probabilities of reward

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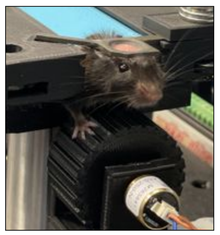
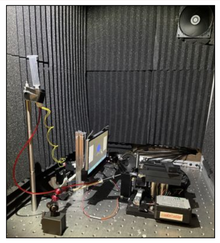
INTRODUCTION

- Reward values dictate the way we make decisions.
- However, the neural circuitry that plays guide decision making is not fully understood.
- Dopamine and the Basal ganglia are hypothesized to play a critical role in reward learning and motivation.
- Here, we designed a head-fixed probabilistic reversal task in mice to assess how mice learn probabilistic reward outcomes.

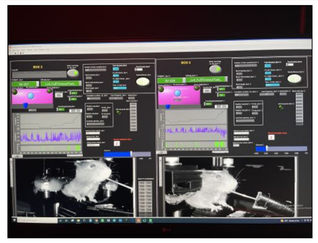
METHODS



3D printed mice chamber



Experimental rig



LabView set up for behavioral task

We ran the behavioral task outlined below on head fixed mice

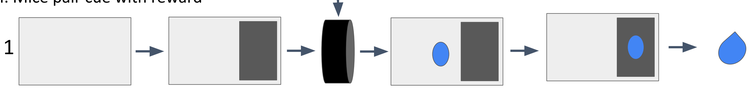
Shaping

Level

Lvl 0

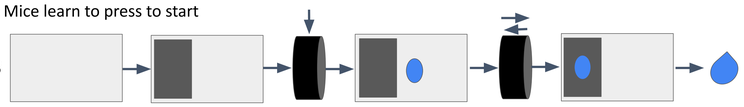
Description: Head fixed mice observed the full task
Goal: Mice pair cue with reward

Lvl 1



Description: Mice have press to start, observed the full task
Goal: Mice learn to press to start

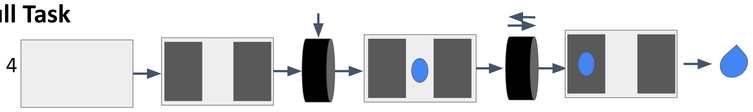
Lvl 3



Description: Mice press down and spin wheel to complete task
Goal: Mice learn they control the ball

Full Task

Lvl 4

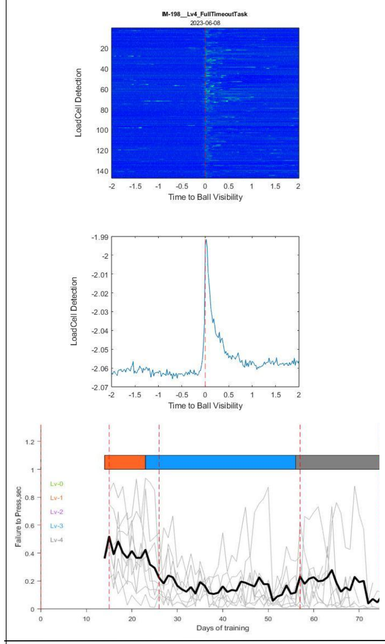


Description: Different probabilities of reward associated with left and right box choice

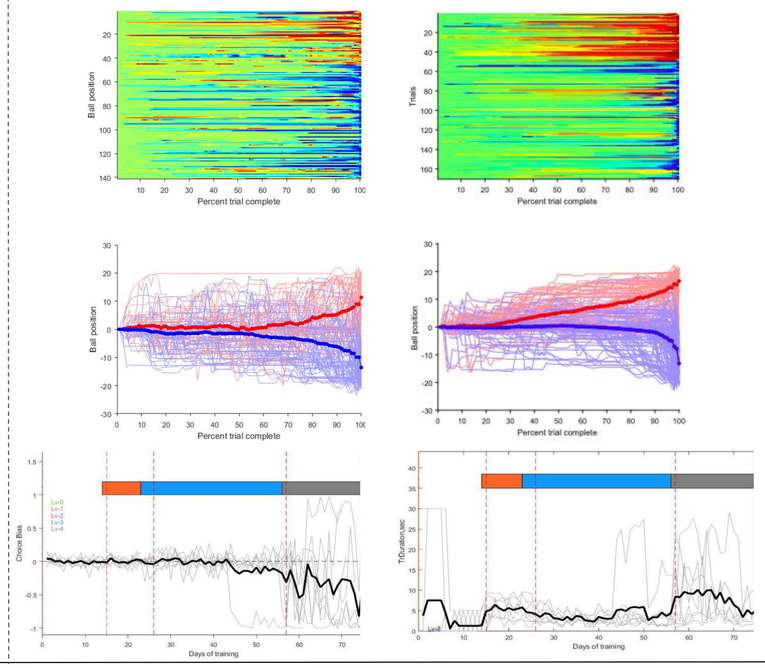
Left Prob	0.1	0.5	0.9	0.1	0.1	0.5	0.5	0.9	0.9
Right Prop	0.1	0.5	0.9	0.5	0.9	0.1	0.9	0.1	0.5

RESULTS

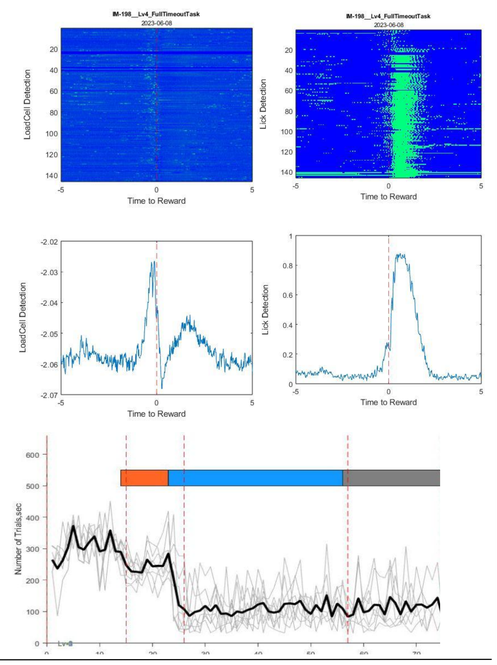
Mice press the wheel to start



Mice Spin the wheel to make a choice

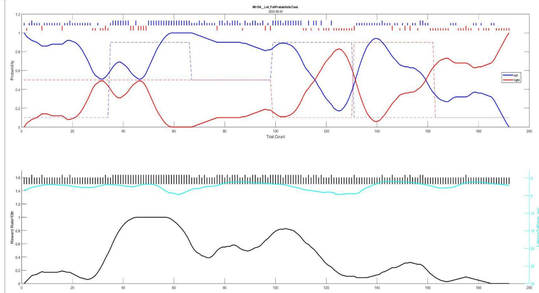


Mice licks for reward

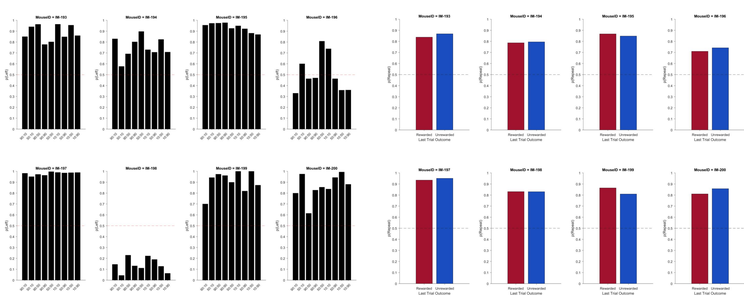


Full Task

Decision making behavior is influenced by reward probability

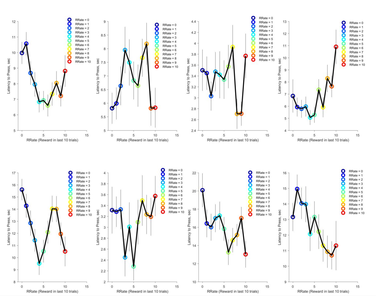


Average mice choice based on reward probability



Probability of repeating reward vs unrewarded trails

Motivation in correlation to reward rate

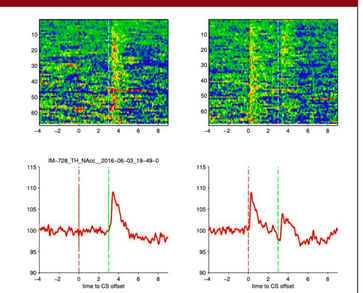


CONCLUSIONS

- We developed a behavioral task that can assess if mice can differentiate more favorable probability of reward.
- Mice are able to learn to press wheel to initiate trial and lick for reward.
- The mice have a strong leftward bias making the full probabilistic task inconclusive in assessing whether they can differentiate favorable probabilities of reward.
- Some mice showed correlation between latency to press and reward rate indicating their actions are goal directed.

FUTURE

- Gather neural data to assess changes in dopamine during these behavioral task to evaluate the role of dopamine in decision making.
- This will be done through the use of photometry and microdialysis.



ACKNOWLEDGEMENTS

Thank you to the MnDrive program, North Star STEM Alliance, and my mentor Dr. Arif Hamid.

This project was supported by the Louis Stokes North Star STEM Alliance, National Science Foundation award number CON000000064472, and the University of Minnesota's MnDRIVE (Minnesota's Discovery, Research, and InnoVation Economy) initiative.

Heterotroph Bacteria coexist with Cyanobacteria: Can Heterotrophic bacteria break down Microcystin?

Sumaya Khalif, Bea Baselga-Cervera & Michael Travisano

Department of Ecology, Evolution and Behavior, University of Minnesota Twin Cities

Introduction

Do you know that Blue-green algae bloom are polluting our freshwater with Microcytin toxin, causing harm to both humans and animals?

- Blue-green algae blooms is an overgrowth of cyanobacteria in warm and still-moving fresh water.
- Some cyanobacteria can produce toxins such as microcystin. Cyanobacteria coexist with heterotroph bacteria.
- Microcystin is a very dangerous liver toxin and potential human carcinogen. This toxin can also cause other health complication and can kill livestock and pets that consume the affected waters.

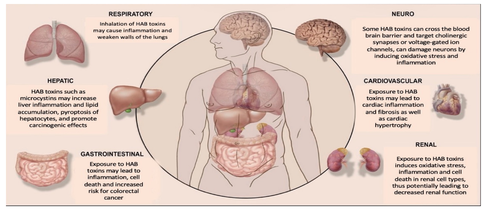


Figure 1. Health complication that can occur from exposure to microcystin.

Main Goals

- Investigate if heterotrophic bacteria break down Microcytin.
- Identify and isolate heterotrophic bacteria strains.

References

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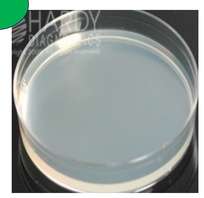
Methodology

A sample was taken from a lake contaminated by blue-green algae bloom in Minnesota.

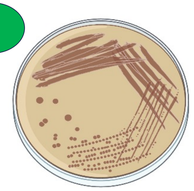
- We used R2A media which is a rich media for freshwater bacteria.
- We isolated heterotroph bacteria from a microcystin producing cyanobacteria culture and used a 16S DNA amplification to identify the heterotrophs.
- Sonicator is used to break down the cells to release the toxics (Spent media).
- We did toxicology and microcystin biodegradation experiments with the isolated heterotrophic bacteria strains.

1



R2A media

2



Isolated single colonies

3

16S DNA amplification

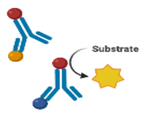
4

Sonicator

5

Tecan – plate readers

6



Microcystin ELISA

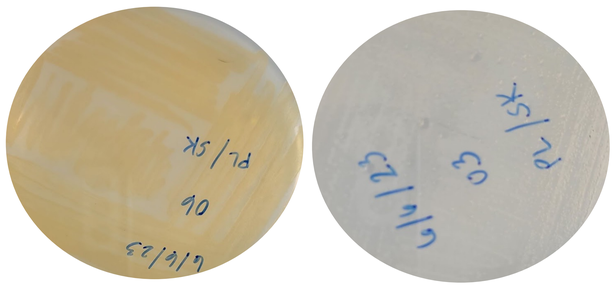


Figure 2. Example of an identified Blastomonas *aquatica* bacterial strain (left) and an unidentified bacterial strain (right).

Results

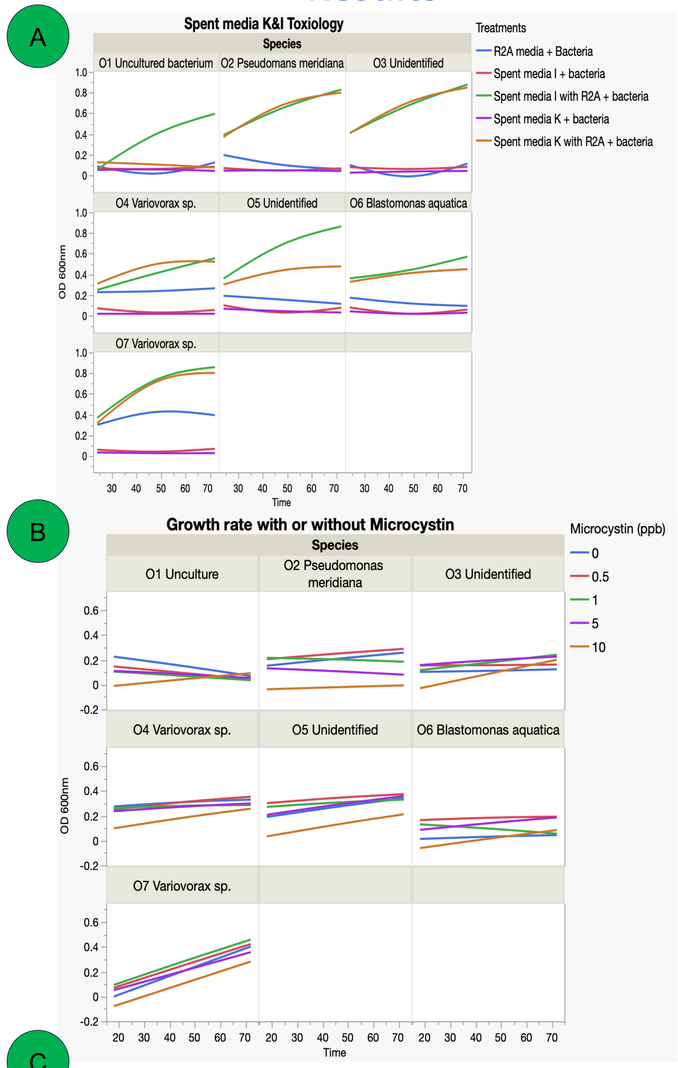


Figure 3. Growth curves of the seven isolated heterotrophs over 72hrs. Strains were cultured in R2A media, and two different spent media enriched and not with R2A. Results indicate that all the heterotrophs grow better in spent media with nutrients (A). 72hrs growth curve of the seven isolated heterotroph species at different doses of microcystin (ppb). We observed a growth inhibition at higher doses in some species (B). Concentration of the Microcytin is very high showing the bacterial stains did not break down microcytin (C).

Conclusion

- Overall, The experiment tells us that after 72 hours of growth, the heterotrophic bacteria strains did not break down the microcystin based on the Elisa biodegradation graph.
- The spent media K&I growth shows that the bacteria grow to higher concentrations with both the spent media and the R2A which can mean that they are using some secondary metabolites produced by the cyanobacteria.

Future directions

- Redoing the ELISA experiment and increasing incubation period.
- Redoing the DNA extraction and sending it to sequencing for the unidentified bacterial strains.

Acknowledgements

Project supported by the Louis Stokes North Star STEM Alliance, National Science Foundation award number CON000000064472, and the University of Minnesota's MnDRIVE (Minnesota's Discovery, Research, and InnoVation Economy) initiative.

Effects of Prenatal Choline Supplementation on the Expression of Synaptic Plasticity Genes on the Formerly Iron-Deficient Adult Female Rat Hippocampus

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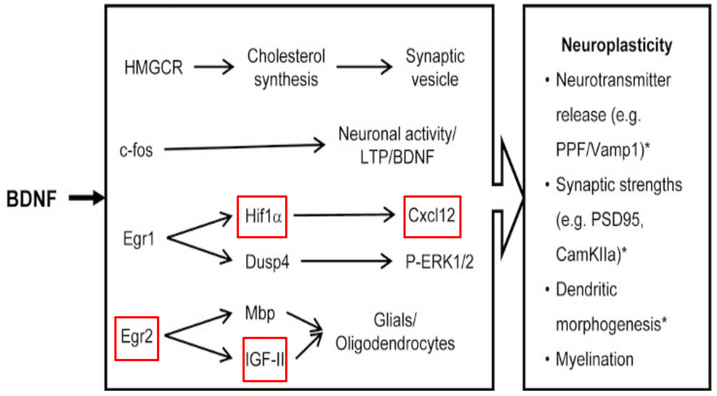
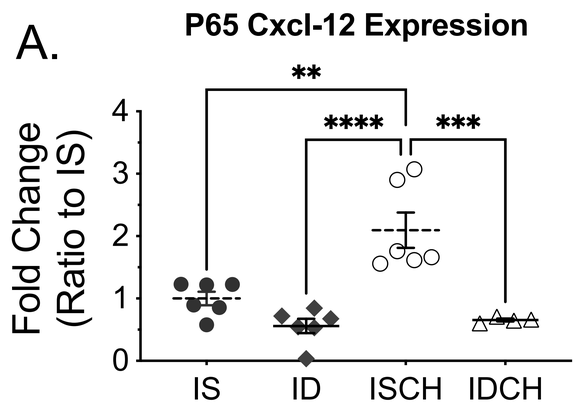
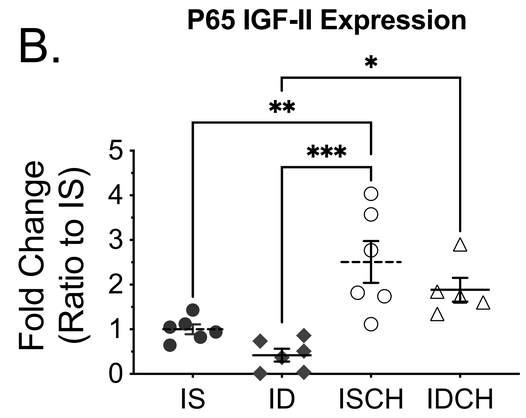
Introduction

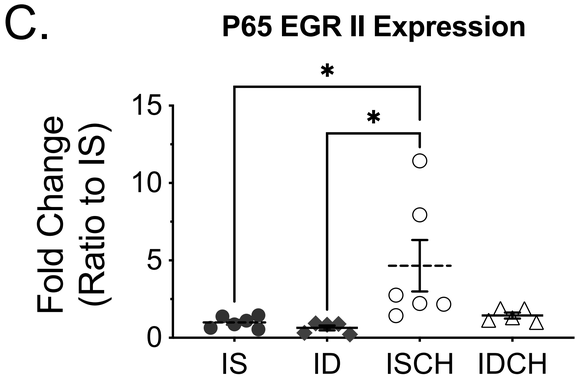
- Iron Deficiency (ID) occurs when there is a chronic reduction of iron intake in an individual's body [3]. This leads to a low number of healthy blood cells, anemia, and symptoms such as extreme fatigue and heart failure [3].
- Pregnant women with ID need more iron due to their high blood volume, putting the baby at risk of ID-associated developmental delays such as slow memory, attention processes, decreased motor activity, and poor learning acquisition [1].
- Choline is a crucial nutrient the brain needs to regulate an individual's memory function [2].
- Prenatal choline supplementation improves neonatal ID-impaired cognitive functioning in the rat model [4].
- **Is there a change in hippocampal gene expression at postnatal day (P) 65 in formerly ID female rats when choline supplementation is given prenatally?**



Results







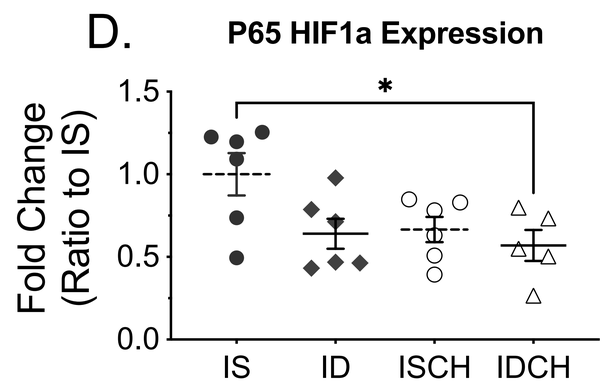
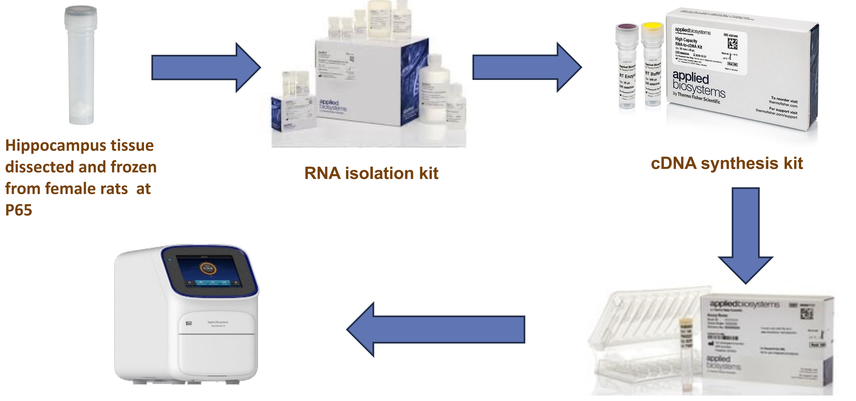


Figure 1. Cxcl-12 plays an important role in the formation of blood cellular components (A). IGF-II contributes heavily to fetal growth and development (B). EGR-II acts as a transcription regulatory factor (C). HIF1a is a head regulator of stabilizing low levels of oxygen in body tissues (D).

Methods

Animal and diet: A diet of either an iron-deficient (4 mg/kg Fe) or iron-sufficient (200 mg/kg Fe) were fed to pregnant rats from gestational (G) day 2 to P7. From G11-18 their diets either did or did not consist of choline supplementation (5 g/kg choline).



Conclusion

- Prenatal choline supplementation's effects on hippocampal gene expression are shown in Figures 1-3 to be more effective in P65 iron sufficient female rats.
- Levels of hippocampal IGF-II expression are shown to increase in ID P65 female rats when treated with prenatal choline supplementation vs. untreated as shown in Figure 2.

Acknowledgments

Project supported by the Louis Stokes North Star STEM Alliance, National Science Foundation award number CON000000064472, and the University of Minnesota's MnDRIVE (Minnesota's Discovery, Research, and InnoVation Economy) initiative.

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Exploring the conditions for crosslinking of a water-soluble polymer when exposed to radicals

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[1]Biology department, Augsburg University; [2]Civil, Environmental and Geo-Engineering Department, University of Minnesota

University of Minnesota
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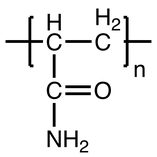
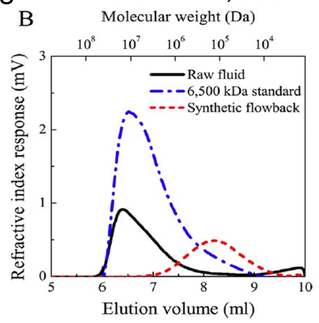
Introduction



Polyacrylamide

Polyacrylamide (PAM) is a water-soluble polymers used as a flocculant agent in water treatment, soil conditioner In agriculture, and a friction reducer in hydraulic fracturing.[1] (95% of acking in the US → 75,000



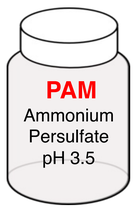
Previous work has shown that when PAM is exposed to radicals, it *decreases in molecular weight*.[2]

Recent results have shown that **under certain conditions PAM will increase in molecular weight** rather than decrease

Objectives:

- Explore the polymer and radical concentration that results in an increase in molecular weight
- Develop a method to explore the solid formed during exposure to radicals and understand the conditions when it forms

Methods



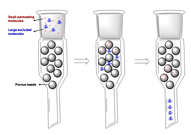
24 hrs

Hot water bath
80°C

Analyzers

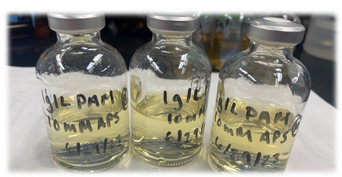
Total Organic Carbon (TOC) analyzer

Measures the amount of dissolved organic carbon

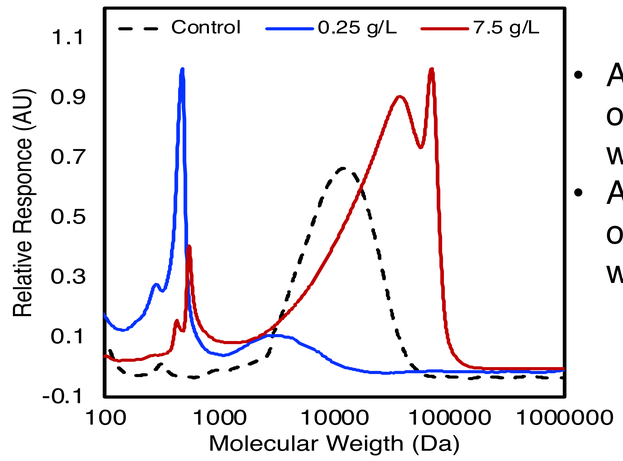


Size Exclusion Chromatography (SEC)

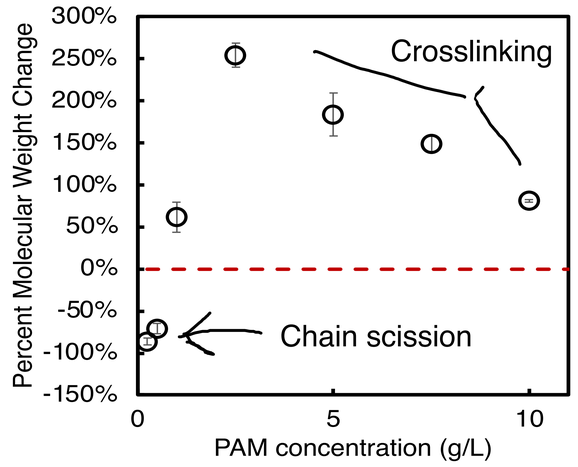
Measures the molecular weight distribution of the polymer



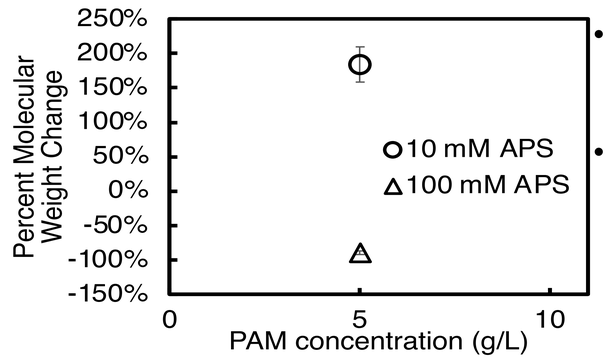
Effect of Polymer and Radical Concentration



- At higher concentration of PAM, molecular weight increase
- At Lower concentration of PAM, molecular weight decrease

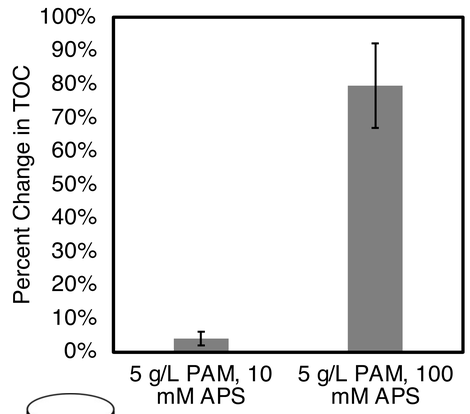


- Dots below red line indicate no change in molecular weight over the degradation period
- Above red line means increase in molecular weight over degradation period

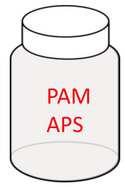


- 10 mM APS shows percentage increase in PAM molecular weight
- 100mM APS shows percentage decrease in PAM molecular weight

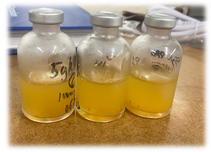
Solid Formation



5g/L Pam
100mM APS



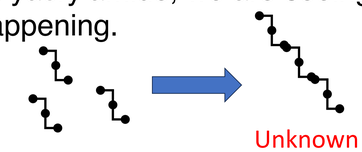
Filter



Oven

Conclusions

- At certain concentrations of polyacrylamide, we are seeing crosslinking happening.



Unknown product

- Understanding the chemical changes of polymer in environmental condition give us information about the fit of the polymer in the environment

ACKNOWLEDGEMENTS

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Designing Pollinator Friendly Seed Mixes for Solar Farm Habitats

Trisha Trinh - University of Minnesota, Twin Cities

BACKGROUND

- Solar fields are becoming popular thus more agricultural areas (corn/soy) are being converted. However, at **what cost is this on the biodiversity and the pollinator habitats?**
- As defined by Forest Service U.S.D.A, **pollinator habitats** consist of native shrubs, grasses and/or wildflowers which provide cover, nectar and pollen for native pollinators such as bees, butterflies, beetles, and moths [1].
- Solar fields can have **different microclimates** that depend on their shade amount, heat radiation, and moisture/humidity which affect the effectiveness of current pollinator seed mixes and habitat management recommendations [2].
- In response to these uncertainties, the Board of Water and Soil Resources (BWSR) has implemented the **Habitat Friendly Solar Program** to access and offer suggestions to solar field owners.

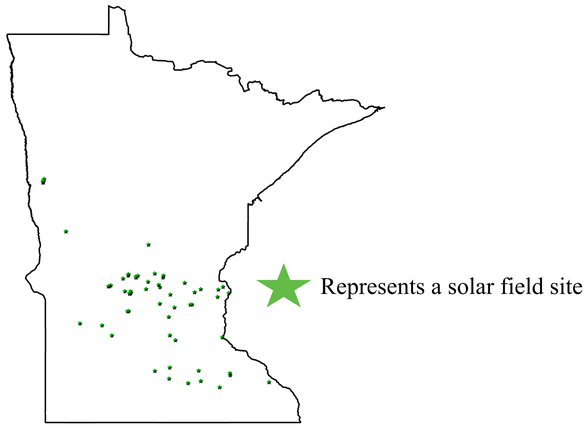


Figure 1: illustrates the solar field locations throughout Minnesota

GOAL

This study will identify which components in pollinator seed mixes and habitat management recommendations are most effective in maintaining and establishing successful pollinator habitats.

OBJECTIVES

1. Analyze vegetation and wildlife in solar field sites throughout Minnesota.
2. Identify which components in pollinator seed mixes and habitat management recommendations are most effective in maintaining and establishing successful pollinator habitats.
3. Determine what environmental factors contribute to the growth and sustainability of pollinator habitats.

METHODS

Field Observation → Assessment surveys in the field
- BWSR Solar Site Assessment Form
- Meandering Surveys

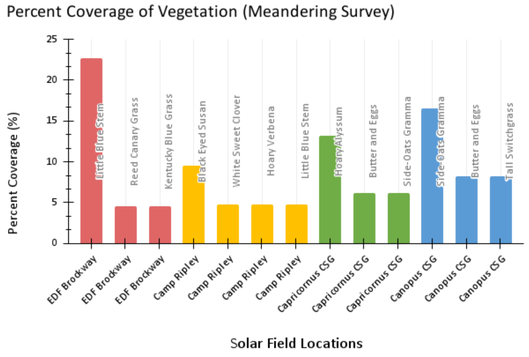
→ Percentage coverage analysis

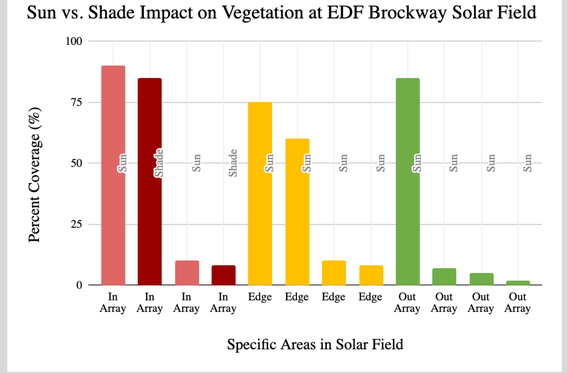
Figure 2: demonstrates collecting soil samples.

Figure 3: highlights the essential meandering data collection process.

Figure 4: shows a bee fly pollinating a plant.

RESULTS





Bee Species at Different Solar Field Locations

Capricornus CSG (total: 28)	Canopus CSG (total: 25)	Camp Ripley (total: 36)	EDF Brockway (total: 27)
• Honeybee (25) • Two-Spotted Bumblebee (1) • Half-Black Bumblebee (2)	• Honeybee (13) • Common Eastern Bumblebee (2) • Brown-Belted Bumblebee (2) • Unknown Bumblebee Species (1) • Unknown Bee Species (7)	• Honeybee (7) • Common Easter Bumblebee (13) • Unknown Bumblebee Species (4) • Brown-Belted Bumblebee (7) • Two-Spotted Bumblebee (1) • Half-Black Bumblebee (1) • Golden Northern Bumblebee (1) • Unknown Digger Bee Species (1) • Unknown Bee Species (1)	• Honeybee (13) • Brown-Belted Bumblebee (3) • Common Eastern Bumblebee (5) • Unknown Bee Species (4) • Black and Gold Bumblebee (2)

DISCUSSION

- Currently preliminary analysis on shaded vs. sunny and in EDF Brockway Solar Field are in progress. However there are much more data to further assist in this study,
- Grasses such as Little Bluestem and Yellow Indian Grass had the most percent coverage.
- Vegetation was more prominent in sunny areas versus shade.
- Camp Ripley was found to have the most bee species during the field study.

CONCLUSION

- Overall, there are different factors that contribute to a solar field's success in supporting a pollinating habitat.
- The shaded vs. sunny areas and different bee species were the main criterias analyzed. However there are most certainly more things to analyze to strengthen the correlations. Other factors such as native vs. invasive species, bloom season, etc. will allow better understanding of the impact of certain seed mixes at solar fields.
- All in all, the there is a multitude of studies that can help local solar field owners better take care of their land and further help increase and strengthen the biodiversity of the pollinating habitats that are crucial to our everyday lives.

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