The Friends of Nachusa Grasslands
2022 Scientific Research Project Grant Report
Due June 30, 2023

Please answer the following questions with clearly written summaries to give Nachusa Friends' science committee members, officers, and board members a good idea of what you accomplished using your grant money. Unless you object to the Friends doing so, your report will be uploaded into the science section of the Friends' website: nachusagrasslands.org. Donors and prospective researchers often read these reports after they are posted.

1. Please save this form to your desktop with a unique file name that includes "Friends 2022 Science Grant Report" and your last name.
2. Complete the form using the headings in bold as your guide.
3. Save the file as a Word document or a PDF.
4. Attach the file to an e-mail, and send it to: nachusafriendsscience@gmail.com no later than June 30, 2023.
5. The subject of the e-mail should be "2022 Scientific Research Grant Report" and your last name.
6. If you have not completed your work, please submit this form anyway by the June 30 deadline and plan to contact Friends after your project is complete so that we may learn from and publicize the outcomes as appropriate.

Name: Laurie Spencer
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2022 grant amount: $2,710

Research Project Topic:

Characterization of Intestinal Microbiome in Wild Bison at Nachusa Grasslands

Research Project Purpose:

The intestinal microbiome aids in digestion and immune system function. Disruption of this microbiome can result in negative health effects. Parasite infections are major threats to bison, particularly in herds contained within smaller, more concentrated environments. Intestinal parasite infections have the potential to cause weight-loss, anemia, and diarrhea and disrupt the gut microbiome, which can strain an animal's immune system. Therefore, my study aims are to characterize to intestinal microbiome

and parasite infections, longitudinally, of the Nachusa bison through various microbiological and molecular methods.

Research Project Outcomes to date:

DNA was extracted from bison fecal samples as the first step in the molecular analyses. A portion of the bison DNA samples were segregated for sequencing and the library preparation process was conducted in our lab as the first step in the sequencing workflow. Upon completion, the samples were shipped to Rush University Microbiome Core Facility (Chicago, IL) where high throughput sequencing of the 16s rRNA gene was performed using an Illumina platform. Upon completion, Rush University electronically shared the sequencing results with our lab, and we then performed bioinformatics analyses using the QIIME2 application. These analyses included assigning taxonomic classifications to the sequence reads, quantifying relative abundances at various taxonomic classification levels and performing statistical tests on the relative abundance differences across the seasons and across the years (2018-2021). We found statistically significant increases in the number of different bacteria and the relative abundance of these taxa from 2018 through 2021. Significant seasonal differences were also noted; the number of different bacterial taxa were highest in spring and summer, the relative abundances were most similar in spring and summer with vastly different relative abundances in fall and winter. Microscopic analysis of bison fecal samples (collected in 2021) revealed 9 different types of intestinal parasites (at the genus level). Based on these results, primer sets (specific to these types of parasites) were purchased for molecular detection of these parasites using PCR. This PCR analysis utilizes DNA extracted from the fecal samples (2018-2021), and currently in process. Preliminary PCR results for 4 different intestinal parasites can be seen in **Figure 1**, below. Upon completion, final parasite results will be shared.

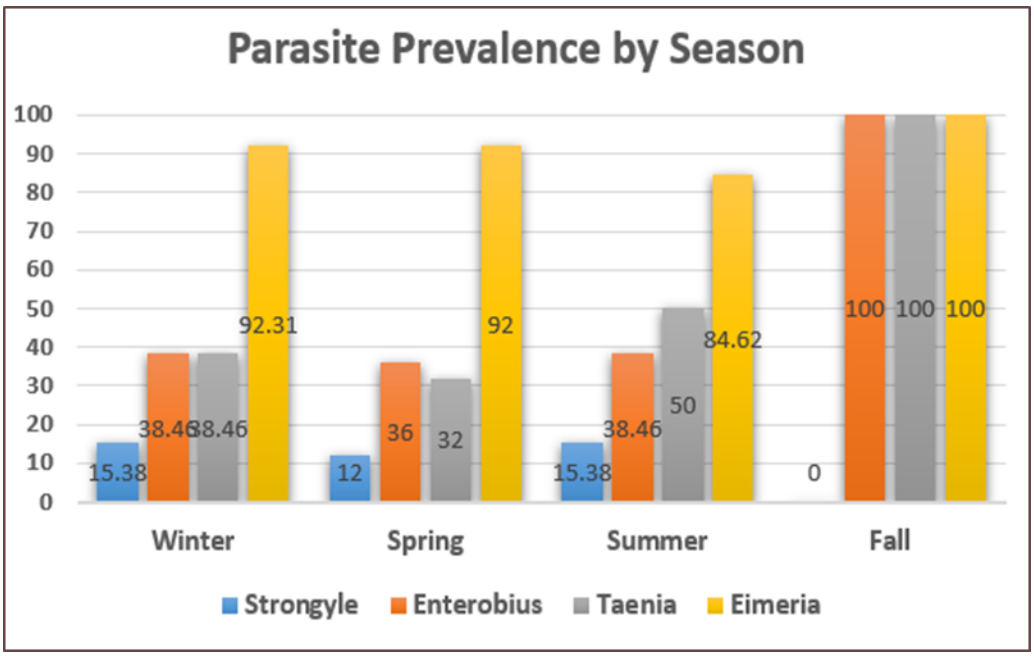

Figure 1. Parasite prevalence (percent infected), by season.

Describe how the grant funds you have received from the Friends of Nachusa Grasslands have been used in regard to the above topic, purpose, and/or outcomes:

These grant funds were utilized to purchase laboratory supplies for the microscopic and molecular analysis of intestinal parasites and bacterial microbiome composition of Nachusa bison. These supplies include DNA isolation kits, PCR primer sets and PCR reagents for parasite detection and for the "library preparation" of DNA samples (prior to DNA sequencing). These funds were also used to purchase staining kits for the microscopic identification of parasites.

Describe how your project has benefited the work and goals of Nachusa Grasslands:

Wild American bison (*Bison bison*) were reintroduced to Nachusa beginning in 2014 as part of the Nature Conservancy's mission to restore native tallgrass prairie. Ensuring the health of these bison is paramount to the sustainability of these ecosystem restoration efforts.

Describe how your findings can be applied to challenges in management practices for restoration effectiveness and species of concern:

Finalized results from this study can be used by the Nachusa Grasslands team to better understand the health of the bison herd. These findings may assist with decisions on veterinary health interventions (e.g., anti-parasitic medications provided during annual round-ups) or other herd management strategies (e.g., preference to certain areas of the preserve for grazing).

Please list presentations/posters you have given on your research:

1. Phi Sigma Symposium at NIU: Poster (April 2022)
2. Nachusa Science Symposium: Talk (April 2022)
3. American Society of Microbiology Conference: Poster (June 2022)
4. NIU Biology Dept. Colloquium Series: Talk (December 2022)
5. Phi Sigma Symposium at NIU: Talk (April 2023)
6. Nachusa Science Symposium: Talk (April 2023)

Have you submitted manuscripts to scientific journals? If so, which ones? If not, do you anticipate doing so? (Please send digital copies of published articles to the Friends so that we can learn from your work.**)**

Not yet, however, I have a manuscript draft in process for this project specific to the results obtained from the bison intestinal microbiome DNA sequencing analysis. Upon completion, this manuscript will be submitted to a relevant peer-reviewed scientific journal for publishing.

What follow-up research work related to this project do you anticipate (if any)?

I will continue performing PCR detection for the remaining parasite types. I will also be conducting additional bioinformatics analyses on the relationships between parasite infections and temperature/precipitation fluctuations on the intestinal microbiome compositional shifts across years and seasons. Future research on this subject will include an expansion of herds from other locations for comparison of results. In addition to testing other conservation herds, analysis of bison from commercial farms would allow for better understanding of factors that may influence the intestinal microbiome composition and parasite transmission dynamics in bison.

<u>Optional</u>: Suggestions for improving the application and award process for future Friends of Nachusa Grasslands Scientific Research Grants: