**The Evergreen State College**

**Graduate Program on the Environment**

### Thesis Prospectus

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**STUDENT AGREEMENT:**

**SIGNATURE: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ DATE\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_**

**FACULTY READER APPROVAL:**

**SIGNATURE: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ DATE\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_**

**MES DIRECTOR APPROVAL:**

**SIGNATURE:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ DATE\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_**

1. **Provide the working title of your thesis[[1]](#footnote-1).**

Cryoconite Holes: implications on downstream ecosystems

1. **In 250 words or less, summarize the key background information needed to understand your research problem and question.**

Cryoconite holes can be found all over the world’s glaciers and ice fields. These holes are created when sediment lands on ice and changes the albedo. This change in the localized area initiates melt and over time a pool of water forms. Within these pools grows a wide variety of life, including sediment, microbial communities, plant matter and fungi. As rapid global glacial melt continues to increase so does the rate of distribution of this microbial life from cryoconite holes into glacial headwaters.

Cryoconite holes are understudied, and little is known about their influence on the downstream carbon cycle. Yet, input from these structures is significant enough to warrant inclusion into models of downstream ecosystem processes. As cryoconite waters enter hydrological systems, influxes of sediment and microbial communities may impact soil development in recently deglaciated areas and downstream systems. These glacial headwaters are the beginning of complex and evolving ecosystem gradients that span from alpine to marine territory. The social and ecological infrastructure that millions of people depend on for their livelihoods depend on these water systems. As we continue to strategize our future within a model of enhanced global warming, the impact of microbial communities within glacial melt is extremely important to understand. Gaining further knowledge on how large influxes of carbon and microbial communities into streams and water systems will help update our current hydrological models and better prepare us for the future ahead.

1. **State your research question(s).**

Does the addition of organic matter from cryoconite holes impact ecosystem respiration rates in glacially fed rivers?

1. **Situate your research problem within the relevant literature. What is the theoretical and/or practical framework of your research problem?**

Glacial melt is responsible for contributing water to downstream ecosystems. These contributions are often the most important throughout seasonally dry months, when glaciers thaw and little water is contributed by other systems. Perennial stream habitats worldwide are created by this ebb and flow of water availability, an essential resource for plants and animals (Telling et al., 2010). By nature, glacially fed watersheds originate in extreme ecosystems where life is spread thinly on both spatial and temporal scales. As a result, glacier runoff is often mostly just water, precipitated, frozen, then thawed and running down and from the icy mass. As climate change accelerates the rate of glacier melt and increases the contributions of exposed and growing cryoconite holes into their nearby hydrological systems, the amount of discharged labile organic matter grows in tandem. The inherently desolate nature of alpine and glaciated ecosystems yields these contributions as potentially extremely important to downstream ecosystem processes and respiration. Cryoconite water is responsible for up to 15% of all glacial meltwater runoff (Fountain et al., 2008). These holes are diverse habitats harboring a range of microbial life and higher-level organisms that are responsible for significant rates of primary and secondary productivity (Hodson et al., 2008). Despite these facts, current hydrological models do not yet recognize the impact and potential outcomes of cryoconite water entering downstream ecosystems at a faster rate than seen before.

1. **Explain the significance of this research problem. Why is this research important? What are the potential contributions of your work? How might your work advance scholarship?**

Isolated and lasting within their icy mass, cryoconite holes are often referred to as “icy hot spots” because of the densely diverse life held within them (Edwards et al., 2013). They cover 4-6% of all glaciated surfaces around the world, often persisting for decades (Fountain et al., 2004, Tranter et al., 2004). Many studies indicate that meltwater inputs from these formations are significant enough to warrant inclusion into models of downstream ecosystem processes (Bagshaw et al., 2013). With glacial melt becoming more rapid, the subsequently increased volume of water released from cryoconite holes could greatly impact downstream ecosystems. While studies have been conducted worldwide on cryoconite holes and their potential impacts, none have assess their impact on downstream respiration rate within the United States. Similarly, I have been unable to find a study that removes organic matter from the cryoconite hole and directly adds it to the glacial fed river below the terminus, potentially highlighting the lability of cryoconite sediment entering downstream systems and increasing respiration rates. This research will contribute to the growing number of studies conducted on glacial ecosystems, adding valuable data.

1. **Summarize your study design[[2]](#footnote-2). If applicable, identify the key variables in your study. What is their relationship to each other? For example, which variables are you considering as independent (explanatory) and dependent (response)?**

I will examine the variance of respiration in two study areas, within cryoconite holes on the glacier and 50 m from the terminus of glacial outflow. Water and sediment samples will be collected and placed in BOD glass bottles. Respiration and photosynthesis will be evaluated within the cryoconite holes after 24 hours. While these incubations are taking place, physicality measurements of cryoconite holes will be taken. Within a chosen area, measurement of distance apart, radius, depth of water, depth of sediment and total holes will be recorded. In addition, samples from randomly sampled holes will be collected and stored for microbial analysis to be conducted at the Evergreen State College. Random sampling of sediment from the cryoconite holes will also be collected and sent out for % Carbon (C) and % Nitrogen (N) analysis. Fifty meters from the glacial terminus, respiration rates will be measured as follows: First, sediment from the cryoconite holes will be added to a BOD bottle, with river water (50 m from terminus) added to it, and bottles will be incubated for 24 hours. Next, respiration rates will be measured from unaltered river water collected 50m from glacial terminus. Water samples will be taken for microbial analysis and % N and % C analysis.

* 1. Independent (explanatory) variable: 1) Location (glacial river water verses cryoconite hole). Also, 2) presence of cryoconite organic matter (for the comparison of stream water w/ and w/out cryoconite) and DOC concentrations.
	2. Dependent (response) variable: Respiration rate

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| *Data Collected* | *Questions addressed* | *Methods used* | *Intended Use* |
| Physical Characteristics of Cryoconite Holes | What is the physicality of cryoconite holes in the study area?  | Measurement of distance apart, depth and total holes in chosen area.  | To understand total amount of cryoconite holes in an area and to collect possible georeferencing data for GIS |
| Respiration and Photosynthesis of Cryoconite Holes | What is the respiration and photosynthetic rate within cryoconite holes? | Closed dark bottle incubations and light bottle incubations on glacier.  | To determine the respiration and photosynthesis measurements within cryoconite holes |
| Collection of Cryoconite Water(Microbial) | What is the abundance of microbial communities within cryoconite holes? | Water will be removed from each hole and later evaluated with microscopy. | To evaluate abundance and basic diversity of microbial communities within the holes  |
| Collection of Cryoconite Water (DOC Concentration mg/L) | Is there a difference of % dissolved organic carbon amongst holes and at the glacial terminus? | Water will be removed from each hole and later evaluated with microscopy.  | To evaluate carbon percentages across hole and in the downstream environment.  |
| Collection of Cryoconite Sediment(C:N ratio, %C and %N) | What is the C:N ratio of the cryoconite sediment within the holes? What is the % C and the % N?  | Sediment will be removed from each hole and evaluated for the C:N ratio %C and % N | To evaluate lability within the cryoconite holes. |
| Respiration of Freshwater at Glacial Terminus | What is the respiration rate 50m from glacial terminus? | Closed dark bottle incubations near terminus. | To determine respiration rate of freshwater downstream from glacial output |
| Glacial Terminus Water Amalgamated with Cryoconite Hole Water | Does the respiration rate of the water from the glacial terminus change when cryoconite hole water is added? | Water from the cryoconite hole added to the BOD bottle with water from the glacial terminus and incubated for 24 hours. | To determine change in respiration rate with added cryoconite hole water. |

1. **Describe the data that will be the foundation of your thesis. Will you use existing data, or gather new data (or both)? Describe the process of acquiring or collecting data[[3]](#footnote-3).**

All samples collected will be assessed for pH, temperature, respiration rate, photosynthesis rate, microbial abundance, Dissolved Organic Carbon (DOC) and potentially % C and %N for the cryoconite material.

*Cryoconite Water Collection*: Respiration and rate of photosynthesis will be measured at three cryoconite holes. Organic Matter (OM) will also be collected. At each chosen hole, cryoconite will be removed using a plastic spoon. This collection will be distributed into six 60 mL BOD glass bottles with a final sediment depth of 2-4 mm. All bottles will then be filled with glacial water from the hole. Using a 60 mL plastic syringe, each bottle will then be filled to the top with *in situ* supraglacial water, leaving no head room. An additional three bottles per hole will be used to assess initial oxygen concentration and filled only with cryoconite hole water. Temperature will be held constant by doing the incubations *in situ*. Oxygen concentrations will be measured in each individual bottle using Winkler titrations. All nine bottles per hole will be closed with a glass stopper, and three containing sediment will be designated as dark bottles and be covered with aluminum foil. The light bottles will be used to aid in determining photosynthesis rates. Bottles will then be placed into their respective cryoconite hole and incubated for 24 +/- 2 hours.

*Physicality Data Collection*: Within the chosen plot, 10 cryoconite holes will be located and recorded. Width and depth of all holes in the plot will be measured and GPS coordinates will be noted for future georeferencing. Distance apart will also be recorded. Data will be collected using ArcGIS Field Data Collector. If possible, drone FLIR imagery and photography will be conducted at the site.

*Dissolved Organic Carbon and Particulate Organic Matter*: DOC samples will be filtered through a 0.7 µm GFF filter into an acid cleaned HDPE bottles and stored on ice to be evaluated at UC Davis.

*Glacial Terminus*: Fifty meters from the glacial terminus, stream water will be collected into 18 BOD glass bottles. Nine of these bottles will be amended with OM and water from the cryoconite hole and nine will just contain stream water. Once temperature and dissolved oxygen has been measured, glass stoppers will be used, dark bottles will be covered with aluminum foil and the bottles will be placed back into the glacial stream for a 24 +/- 2-hour incubation.

*Microbial Community Analysis*: Water collected from each hole and 50m from the glacial terminus will be evaluated for microbial abundance using microscopy. Using sterile 50ml syringes, samples will be filtered through a black 0.2 μm pore-size polycarbonate filters and fixed using formaldehyde or paraformaldehyde. These filters will then be frozen and stored for transport. Once back at the Evergreen State College, the filters will be stained using DAPI or SYBR green stain and cell abundance will be evaluated using epifluorescent microscopy. For each filter, the number of cells will be counted in 20 randomly selected fields.

1. **Summarize your methods of data analysis. If applicable, discuss specific techniques that you will use to understand the relationships between variables (e.g., interview coding, cost-benefit analysis, specific statistical analyses, spatial analysis) and the steps and tools (e.g., lab equipment, software) that you will take to complete your analyses.**

Data will be analyzed using an R-package. I will evaluate the two sites for differences in respiration as well as looking for a change in respiration with the cryoconite sample that is added to the downstream sample. The % dissolved organic carbon will also be evaluated. Statistical analysis will be used for normality and variance. Comparisons between sites will be done using a t-test and Pearson’s R will be used for correlation. I will use Principal Component Analysis (PCA) or Non-Metric Multidimensional Scaling (NMDS) to plot how closely related communities within each cryoconite hole are. I will also possibly compare the cryoconite microbial community to the freshwater microbial community.

1. **Address the ethical issues[[4]](#footnote-4) raised by your thesis work. Include issues such as risks to anyone involved in the research, as well as specific people or groups that might benefit from or be harmed by your thesis work, perhaps depending on your results. List any specific reviews you must complete first (e.g., Human Subjects Review or Animal Use Protocol Form).**

To successfully complete my thesis, I will need to make sure I can collect data from Matanuska Glacier. There is no group of people who will be negatively impacted by my research as it focuses solely on glacial runoff and downstream ecosystems and not on people impacted by increased melt itself. While I do recognize that many marginalized communities worldwide are hugely reliant on glacial ecosystems, those topics will not be covered in my outlined thesis.

 There are inherent risks when working within glacial and mountain ecosystems. These risks include variable weather conditions, crevasses, avalanche danger and uncertain walking grounds. Variables and hazards must be considered when creating a safety plan. I have years of backcountry experience as well as two avalanche certifications: AIARE 1 and 2. I was a backcountry EMT for nearly 5 years and have been on glaciers on four different continents. In addition, I have solo summited three high altitude mountains including Stok Kangri in Northern India which is over 6000 meters tall.

 Matanuska Glacier in Alaska is a very accessible, doable trek with ratings on all trails. It is frequented by the general population, with easily identifiable hazards. Despite the ease of this trek, before I go, I will be taking a crevasse rescue certification class through the Alpine Institute in May. I have all the necessary gear to ensure a safe climb including crampons, ice axes, probes and avalanche beacons. Solo travel on glaciers is dangerous, for this reason I will be traveling with a companion who has his AIARE 1 and will also be attending the certification class with the Alpine Institute.

1. **List specific research permits[[5]](#footnote-5) or permissions you need to obtain before you begin collecting data (e.g. landowner permissions, agency permits).**

Matanuska Glacier located an hour and a half northeast of Anchorage Alaska is mostly on private land. In order to conduct research here I will need to confirm with the landowners that I have their permission.

1. **Reflect on how your positionality as a researcher could affect your results and how you will account for this in the research process[[6]](#footnote-6).**

I am a mountaineer who has traveled to multiple countries and have seen first-hand the impacts of rapid glacial melt on marginalized communities. This means I have an emotional and moral connection to glacial melt and the ecosystems it disrupts. While my topic is not focused on human exposure and impact, it could still potentially hold weight with how I view this study.

1. **Provide at least a rough estimate of the costs associated with conducting your research.  Provide details about each budget item so that the breakdown of the final cost is clear**.

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| **Description** | **Cost** | **Quantity** | **Total** |
| Flight | $350/person | 2 | $700 |
| Car Rental | $150/day | 3 | $450 |
| Gas | $0.56 | 614 | $343.84 |
| 60 mL BOD Bottle | $717.83/case | 36 | $717.83 |
| Black Filter (0.22 um) | $103.50/100 pc | 1 | $103.50 |
| % C Testing UC Davis | $28/test $30 setup fee | 7 | $226 |
|  |  | **TOTAL** | **$2541.17** |

1. **Provide a detailed working outline of your thesis.**

  **Chapter One: *Introduction***

1. General introduction of cryoconite holes and melting glaciers
2. Research question
3. Reasons and need for research question
4. Defining basic key terminology, glacial structure and cryoconite hole diversity and creation
5. Importance of research question within greater area of glaciology and global implications

 **Chapter Two: *Literature Review***

1. Brief overview of implications of increase cryoconite hole water into glacial runoff
2. Brief overview of literature reviewed
3. Study model and foundational methods through literature review
4. Hypothesis and justifications
5. Scope of study

 **Chapter Three: *Methods Section***

1. Introduction to study area, method and design
2. In-depth description of study design including materials to be used
3. Explanation of definitions, measurements
4. Description of technique
5. Assumptions of study methods

 **Chapter Four: *Results***

1. Brief overview
2. Findings of study
3. Descriptive analysis
4. Statistical analysis
5. Describe hypothesis and exact data (no opinion)

 **Chapter Five: *Discussion***

1. Full discussion of results and their implications
2. Full discussion of statistical analysis
3. Discussion of unplanned or unexpected occurrences or ways to improve data collection in future studies
4. Full discussion of hypothesis and results
5. Implications of results

 **Chapter Six: *Conclusion***

1. Summary of study
2. Refer to literature review
3. Implications of study
4. Limitations of research
5. Recommendations for future studies
6. Tie back into broader picture and global impact

 **Chapter Seven: *References***

1. **Provide a specific work plan and a timeline for each of the major tasks in the work plan. Be as realistic as you can, even though you will probably need to alter this schedule as you complete the tasks. Remember that faculty readers take time to return your drafts and that the final polishing and formatting of your thesis for binding will take longer than you ever imagined.**

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| **Activity** | **Month** | **Deadline** |
| Lit Review: Cryoconite Holes | November | 18-Nov-21 |
| Prospectus  | November | 2-Dec-21 |
| Prospectus to MES Director | December | 10-Dec-21 |
| MES Thesis Grant Application  | December | 8-Jan-22 |
| Methods: Respiration | January  | 14-Jan-22 |
| Field Data Collector Design | January  | 21-Jan-22 |
| Methods: Microbial Communities | January  | 28-Jan-22 |
| Introduction: Cryoconite Holes | February  | 11-Feb-22 |
| Introduction: Glacial Ecosystems | February  | 18-Feb-22 |
| Revise Methods | February  | 25-Feb-22 |
| Revise Literature Review  | March | 4-Mar-22 |
| Data Collection | June-Jul |  |
| Analysis of Microbial Communities | July | 15-Jul-22 |
| Analysis of Respiration Rates | July | 31-Jul-22 |
| Results  | August-Sept | 30-Sep-22 |

1. **Who, beyond your MES faculty reader, will support your thesis? Indicate support both within and outside of Evergreen. Be specific about who they are and in what capacity they will support your thesis. If you are working with an outside agency or expert, be specific about their expectations for your data analysis or publication of results.**

I am hoping to have support from a former professor at Evergreen, Peter Impara, with safely navigating glacial landscapes. I also hope to have support from my faculty John Kirkpatrick and Mike Ruth.

1. **List the 3-5 most important references you have used to identify the specific questions and context of your topic, help with issues of research design and analysis, and/or provide a basis for interpretation. For each annotated reference, explain how your project specifically connects to the source by extending, challenging, or responding to the conclusions, methods, or implications. For any other sources cited in this document provide a complete bibliographic citation.**

**Bagshaw, E. A., Tranter, M., Fountain, A. G., Welch, K., Basagic, H. J., & Lyons, W. B. (2013). Do cryoconite holes have the potential to be significant sources of C, N, and P to downstream depauperate ecosystems of Taylor Valley, Antarctica?. Arctic, Antarctic, and alpine research, 45(4), 440-454.**

Downstream ecosystems were evaluated for C, N and P levels in the Taylor Valley of Antarctica in this study. Higher concentrations of C, N and P were found in the more biologically productive areas, with cryoconite hole biogeochemical characteristics affected by the position of the glacier within the landscape. The impact of dissolved nutrients and particulate matter stored in the water columns of the holes were found to be released into downstream ecosystems. These particles travel through ephemeral streams and seasonal lakes covered with ice through various hydrological connections. The authors suggest that cryoconite holes are an integral part of the ecosystems and need to be included within models of downstream biological processes. My study would extend this by testing the implications of the article.

**Edwards, A., Douglas, B., Anesio, A. M., Rassner, S. M., Irvine-Fynn, T. D., Sattler, B., & Griffith, G. W. (2013). A distinctive fungal community inhabiting cryoconite holes on glaciers in Svalbard. fungal ecology, 6(2), 168-176**

This paper discusses the basics of cryoconite hole creation and the frequency and abundance of fungus within the holes. Fungal communities were compared from three different high arctic glaciers to nearby tundra and moraine soils using statistical analysis of terminal-restriction fragment length polymorphism (T-RFLP) profiles and culturing. It was found that sediments in cryoconite holes were influenced by factors on each glacier. Mentioned in the article was the previous observation of the importance of filamentous fungi as decomposers in the cryoconite saprotrophic community. This article is relevant to my study because it shows the possible diversity contained within each hole. It also suggests that these holes can be different depending on positionality on a glacier. My study will increase the view of this one by looking at the impacts of microbial communities on decomposition.

**Fountain, A. G., Tranter, M., Nylen, T. H., Lewis, K. J., & Mueller, D. R. (2004). Evolution of cryoconite holes and their contribution to meltwater runoff from glaciers in the McMurdo Dry Valleys, Antarctica. Journal of Glaciology, 50(168), 35-45.**

This paper discusses the evolution of cryoconite holes and how they influence downstream ecosystems. Size, variation and frequency are analyzed along with connectiveness to hydrologic systems. The article explores cryoconite hole water levels and the independent status of each hole at the McMurdo Dry Valleys. The article suggests that roughly 13% of all observed runoff at this glacier is from water in cryoconite holes. This article is important because it gives a rough estimate of how much of glacial runoff is compromised of cryoconite water.

**Telling, J., Anesio, A. M., Hawkings, J., Tranter, M., Wadham, J. L., Hodson, A. J., ... & Yallop, M. L. (2010). Measuring rates of gross photosynthesis and net community production in cryoconite holes: a comparison of field methods. Annals of Glaciology, 51(56), 153-162.**

**Tranter, M., Fountain, A. G., Fritsen, C. H., Berry Lyons, W., Priscu, J. C., Statham, P. J., & Welch, K. A. (2004). Extreme hydrochemical conditions in natural microcosms entombed within Antarctic ice. *Hydrological Processes*, *18*(2), 379-387.**

1. You are not locked into this title; its purpose is to help you identify the main point or topic of your thesis at an early stage. [↑](#footnote-ref-1)
2. You might discuss selection of case studies, sampling methods, experimental design, and/or specific hypotheses you will test. You should also address any specialized knowledge or skills that are necessary to complete the research. [↑](#footnote-ref-2)
3. If you are planning to use existing data, explain the specific source, contact information, arrangement with collaborating agencies, and expectations about use of data and final products of your research. If you are planning to gather new data, describe specific methods, time, place, and equipment that will be required. [↑](#footnote-ref-3)
4. If you’re not sure where to start, consult a ‘Code of Ethics’ or other similar document from an academic society in an applicable field of study. [↑](#footnote-ref-4)
5. If you are collecting ANY samples or data, even observational data, on public lands (city, county, state and/or federal) it is your responsibility to find out the permit requirements BEFORE you collect data. Conducting research with tribal members/on tribal lands will have different and additional requirements. [↑](#footnote-ref-5)
6. Your *positionality as a researcher* refers to the fact that one’s “…beliefs, values systems, and moral stances are as fundamentally present and inseparable from the research process as [one]’s physical, virtual, or metaphorical presence when facilitating, participating and/or leading the research project…” (The Weingarten Blog 2017). [↑](#footnote-ref-6)