A review of trends and stressors related to *Hexagenia* mayfly populations to determine future research development

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ABSTRACT

Mayflies (Order Ephemeroptera) are aquatic insects that play a vital role in maintaining river health and are an important part of aquatic and terrestrial food chains. Recent evidence suggests that mayfly populations are in decline, however, there remains little consensus on what environmental factors may be driving population dynamics. We reviewed existing literature (173 articles) using Web of Science on population trends and stressors of *Hexagenia* mayflies to explore what is known and to identify research gaps. The majority of the articles indicated declining mayfly populations, due to a combination of stressors. XX % of were focused on water temperature. However, several other stressors, such as toxic and salinity, were linked to mayfly declines though there were few articles that found direct causal evidence. Surprisingly few articles included research involving mayflies in the surrounding Great Lakes and the Mississippi River. Therefore, more research should test the effect of increasing water temperatures on mayfly growth and emergence. This research is important considering global change and sensitivity of *Hexagenia* to increasing water temperature. There remains little consensus on which environmental factors drive mayfly population trends, leaving opportunities for future research in the field and laboratory setting.

INTRODUCTION

Rivers provide critical habitat and food resources for wildlife. Rivers also provide numerous ecosystem functions for humans, including flood control, recreation, transportation, irrigation, power generation and food production (Kammerer, 1987). However, human activities have altered the structure and function of rivers by changing river flow regimes, causing pollution, and introducing non-native species into the waters (Karr and Chu, 1999). Anthropogenic climate change and associated widespread climate warming threatens rivers and sensitive riverine biota by inducing numerous stressors for aquatic organisms. Therefore, identifying the different stressors and their impact of aquatic organisms has become a priority of ecological research and river management.

Rivers are home to diverse communities of aquatic insects and other macroinvertebrates. Aquatic insects are indicators of water quality and ecosystem health because they are sensitive ecological changes within their habitats (Luiza-Andrade, 2020, Jooste 2020). For example, many aquatic insects, such as mayflies, depend on fast water currents that rivers provide and high levels of dissolved oxygen (Klubertanz, 2016). Alterations of river flow regimes and water quality often reduce river flow and lower dissolved oxygen and thus can have a negative effect on mayfly populations (Klubertanz, 2016). Without mayflies and other aquatic insects, riverine food webs may collapse considering insects serve as an important food resource for other invertebrates, fish, and birds. Furthermore, river ecosystem functions, such as nutrient processing and improving water quality, may also be negatively affected by the loss of aquatic insects considering these organisms help process organic matter and collect and filter fine particles of nutrients and sediments from the river (Klubertanz, 2016). This has led to the emergence of interest of global changes in aquatic insects and their habitats through quantitative monitoring to look for any significant trends (Stepanian et al., 2020).

With drastic environmental changes happening more frequently the increase in 'ecological surprises' continues to be affecting freshwater biota more than we realize (Jackson et al., 2015). Marine environments have continuously been hit with stressors such as rising temperatures, biological invasions, habitat destruction by humans, and ecological degradation (Jackson et al., 2015). While the net impact of all these stressors together can be greater than or equal to the net affect of just one single effect, there is potential for adaptation of organisms to stressors that will limit overall change in the ecosystems (Jackson et al., 2015). However, it is not known if and how organisms can

adapt and what the determining factors are that can cause changes in important aquatic insect species.

Mayflies, *Hexagenia*, are large burrowing insects that can be found in larger bodies of water such as the Mississippi River and Lake Michigan and Superior (Klubertanz, 2016). *Hexagenia* are detritivores that help prevent the buildup of biomasses like algae and detritus in rivers and lakes (Kammerer, 1987). Mayflies are an important food resource in that their eggs are eaten by snails and other insect larvae and the nymphs and adults are preyed on by frogs, fish, birds, water beetles and other predatory insects (Salles, 2000). Despite their short-lived life span (1-2 years), and small body size (3 mm to 4 cm in length) many individuals resist predation and serve as good indicators of water pollution (Salles, 2000). The mass emergence of these adult *Hexagenia* mayflies MENTION THE IMPRESSIVE SIZE OF THESE EVENTS IN THE UMR (SEE STEPANIAN PAPER FOR NUMERIC ESTIMATES). Emergence events can be influenced by factors such as changes in competition and growth in both species and predation effects (Corkum, 2010). Mayflies serve an important role in the food chain but this importance will depend on environmental conditions in their habitats.

Recent evidence suggests *Hexagenia* populations have dropped 52% in the Upper Mississippi River area from the years 2012 to 2019, which creates serious concern for what is to come given global change (Stepanian et al., 2020, Figure 1). The causes to this drastic decline could be due to rising water temperatures within the Great Lakes from global warming and rising pesticide use around the waterways (Stepanian et al., 2020). However, the lack of consensus on the determining factors of *Hexagenia* populations leaves an important gap in our understanding of mayfly presence and emergence in river ecosystems.

In this paper, I performed a systematic literature review on *Hexagenia* mayflies to identify the key components of research on these invertebrates and identify research gaps. My literature review consists of two specific goals:

- 1. Discuss ways that research has been conducted in order to recognize unknown or minimally researched stressors of the *Hexagenia* population.
- 2. Identify ideas or suggestions for future research of Hexagenia mayflies.

METHODS

A Review of Literature on Hexagenia mayflies

To begin, I searched terms and words related to the stressors of *Hexagenia* mayflies using Web of Science (date of search 10, 14, 16 December 2020) to locate related articles around the subject (see Table 1). Next, I screened the articles I found by reading the titles and abstracts to identify the relavence and focus of the article. The articles were not counted for and disregarded if there was no mention of *Hexagenia* mayflies being experimented on specifically, either being on the of the experimental test subjects or the main test subject throughout the experimental design. I did not exclude any articles based on the location of research conducted, the experimental design or date of sampling for research. The relevance of the articles location is discussed further on in the review. I retained 173 out of 297 (58.2%) of articles for the content of the literature review based off these criteria found.

Field	Search Term
Торіс	"Hexagenia" AND "temperature"
Topic	"Hexagenia" AND "population"
Topic	"Hexagenia" AND "decline"
Topic	"Hexagenia" AND "stressors"
Topic	"Hexagenia" AND "toxic"
Topic	"Hexagenia" AND "pollution"
Topic	"Hexagenia" AND "growth"
Topic	"Hexagenia" AND "emergence"
Topic	"Hexagenia" AND "salinity"
Topic	"Hexagenia" AND "chemical"

Table 1. Search terms used to compile literature for papers about Hexagenia mayflies.

RESULTS

Evidence of change in Hexagenia mayfly populations

Research has been conducted throughout the early 1900s to the present day to try and figure out the general trend of *Hexagenia* mayfly populations and determine what is the cause of these fluid changes. Giberson et al. (1991) found that after a ten year long survey of the *Hexagenia* population in Southern Indian Lake, Manitoba Canada, that Hexagenia populations followed their predicted outcomes. The populations of these mayflies decreased greatly after the first five years of surveying and then began to slightly increase (Figure 1, Giberson et al., 1991). Swarms of *Hexagenia* mayflies, visible by radar imageryt, may cover the Mississippi River and nearby land during the hot and humid summer months (Stepanian et al. 2019). Stepanian et al. (2019) identified consistent decline in *Hexagenia* population from 2012-2019. They suggested stressors such as higher temperature surface waters and toxins being released from algae blooms that ultimately led to a decrease in oxygen rates found in their habitats and are concerning for freshwater ecosystems.

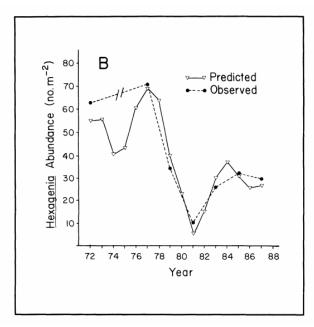


Figure 1. Evidence of predicted and observed Hexagenia populations based on surveys conducted every two years during course of experiment (adapted from Giberson et al. 1991).

While research in Canada show varying results on population trends. For example, looking at Lake Erie as a testing spot for Hexagenia mayflies showed prior to the 1950s these burrowing mayflies dominated the communities of benthic invertebrates of western Lake Erie (Madenjian et al., 1998) The population however saw a large decline due to various reasons and continued to remain very low until 1991 (Madenjian, et al., 1998) From 1991 to 1995 the Hexagenia population in western Lake Erie quickly spread, and looking at regression population models showed a growth rate of 0.92 yr⁻¹, with the conclusion that the once close to extinct *Hexagenia* would have a full recovery rate by 1999 (Madenjian et al., 1998). A new source of observation that has been able to detect the trends in mayfly populations is through radar surveillance. Surveys taken through weather surveillance cameras have been able to investigate more of the trends and patterns alongside waterways such as the Mississippi River and Great Lakes to get better statistical mayfly emergence. The total biomass can be calculated using these radar systems and in recent studies has been exceeded more than 115 billion individuals in the western basin of Lake Erie (Stepanian et al., 2020). Stepanian et al. (2020) also concluded that Lake Erie's western basin population of Hexagenia has seen acute fluctuations that have resulted in a drastic 84% decrease in nymphs. Although these results shed light what is happening to the Hexagenia populations, they do not identify specific factors that could be causing population declines.. Looking at more recent studies have shown however that mayfly population of Hexagenia have declined by over 50% in the Great Lakes region (Stepanian et al., 2020).

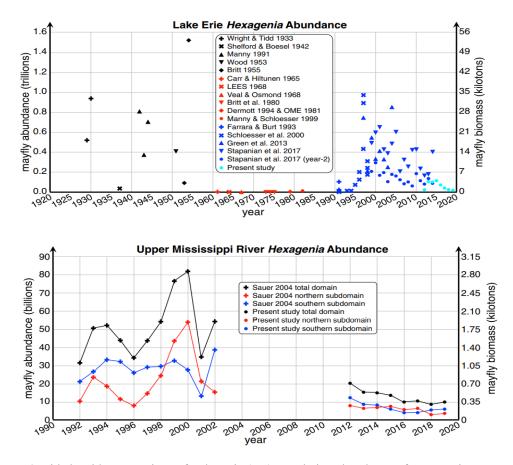


Figure 2. Side by side comparison of Lake Erie (top) population abundance of Hexagenia to Upper Mississippi River (bottom) with the inclusion of other studies and their results indicating the population outcomes corresponding with the year (adapted from Stepanian et al., 2020).

Evidence of causes of mayfly population decline

There were serveral potential causes of *Hexagenia* population identified from the literature (Fig. 3). The most revelant search terms associated with *Hexagenia* population declines were increasing temperature (43%), pollution (19%) and chemical causes (19%).

Looking closer at a revelant article centered to toxins, such as naphthenic acids and metallic naphthenates in a 10:1 mixture of the OSPW contents (Howland et al., 2019). A Canadian study focused on the affects of these concentrations through an acute toxicity test on *Hexagenia* mayflies (Howland et al., 2019). The samples used in this study were found to be both homogeneous and compareable to the Canadian OSPW, or wastewater containing a mixture of dissolved salts, trace metals, and organic compounds, in the ponds of Canada's oil sands region (Howland et al., 2019). The various con entrations of these mixtures lead to the *Hexagenia* test subjects to be immobilized and with increasing concentrations lead to elimination of the whole group of mayflies (Howland et al., 2019). This suggests an issue for *Hexagenia* and other benthic macroinvertebrate communities that live nearby these sites as they continue to make a huge factor in the population decline of the *Hexagenia* mayflies in particular.

Other factors leading to *Hexagenia* declines included the addition of azo dyes to mayfly communities (search word Hexagenia+ toxic), sediment sampling of harbor influenced by urban runoff and waste from steel industries through bioassays (Hexagenia + stressors), eutrophication associated with cyanobacterial toxins and microcystins

(Hexagenia+ pollution).

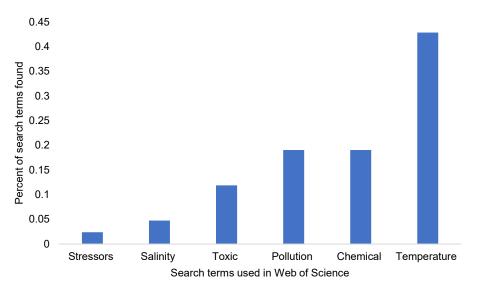


Figure 3. Search terms from Web of Science out of 84 total relevant search terms describing factors describing trends of *Hexagenia* population.

While the search term temperature was the highest percent of relevant causes as to *Hexagenia* decline in populations and individual examination, with over 36 articles relating to or incorporating temperature and its effects within experiments and reviews. Articles regarding delayed egg hatching (Bustos and Corkum, 2013), lake eutrophication involving phosphorous concentrations (Chaffin and Kane, 2010) and peak swarming in *Hexagenia* adults were among some that were defined within the search results. The majority of the search results, however, were not directly centered around temperature being the manipulated variable, but more so a topic of interest due to another variable (Stepanian et al., 2020, Bustos, 2013).

Stressors acting on Hexagenia population

Location of the search article for the stressors found to cause *Hexagenia* decline varied among regions in both the US and Canada, mostly. Out of the 84 relevant articles associated with *Hexagenia* mayfly trends, I specifically looked in the most articles according to the search topics with the trends which was temperature. Out of the 36 articles on the topic of mayfly decline involving temperature, 17/36 took place in the US (Figure 4). Following that, 11 out of the 17 were focused on mayfly population on Lake Erie, others included the Huron River, the Mississippi River and Lakes Huron and Michigan. Surprisingly, 8 out of 36 articles were based on research in Canada with the majority of the research taking place near lakes around Manitoba and Ontario. The other ten articles did not specify where their research took place.

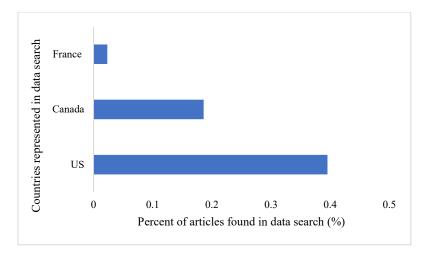


Figure 4. The percentage of articles within the *Hexagenia*+temperature search on Web of Science correlated with what country the experiment specifically took place in.

DISCUSSION

The search on *Hexagenia* mayflies accumulated over 297 articles that used this mayfly genus as a focal point for their research article. Over 58% of those total articles centered their research around the *Hexagenia*. Many of these articles identified different stressors that influence mayfly populations (Table 1). The most definite articles pertaining to those stressors that were searched consisted of temperature (43%), toxins (11%), pollution (12%) and chemicals (12%). Unfortunately, not all articles directly described *Hexagenia* specifically but rather mentioned them as a variable for another organism they were trying to study (fish, other invertebrates, and mussels).

Out of the 43 search topics researched in terms of *Hexagenia* mayfly population, the leading country that had the most articles was the United States. Lake Erie, specifically the western basin consisted of 17/43 of the articles, whereas the other Great Lakes lacked the number of articles including mayflies. This pattern of location followed for the majority of the search terms, with temperature having the largest quantity. Therefore, future research would ideally study *Hexagenia* from not only the other Great Lakes but also the Mississippi River due it's large population of mayflies during the summer months, and its popularity regarding the masses of mayflies that have caused concern with the public (Klubertanz, 2016).

In recent years, temperatures around the Midwest have been unpredictable and varying depending on the season. The National Weather Service has generated future outlooks of temperature and predict that temperatures over the majority of Wisconsin will be 33% higher than average for the months of May through June and a precipitation will be 40% above average (NWS, 2020). Based on my literature review, these predictions could mean the further decrease in the growth and emergence of aquatic insects due to warming water temperature and chemical runoff during flooding. For example, studies showed increasing temperature impacts a delayed hatching response of *Hexagenia* larvae as well as there being an increase in the levels of toxicity from chemicals in their environments (Bustos and Corkum, 2013, Howland et al., 2019). These past studies on the response of riverine biota help guide future experiments for how to manage our environments properly for the species living within them. However, there is still much to be researched for other important species, such as mayflies, that can be conducted including behavior, predator versus prey manipulations, competition rates and size rates. Key research foci for future inquiry could include the manipulation of different water temperatures in both the surface and deeper water levels.

CONCLUSION

By understanding the trends in research of *Hexagenia* mayflies we can identify future research to be conducted from recognizing stressors that have minimal information in regard to the *Hexagenia* populations. It can also ensure that there is research that is done to compare with the articles already published but also to spark new ideas for experiments to take place. *Hexagenia* mayflies are currently on the decline within their populations and we need to consider the factors that are limiting these numbers. The stressors, temperature, salinity, chemicals, toxins, and pollution are some of the most commonly identified factors when describing *Hexagenia* populations. This can be the starting point of future research and how relationships between the terms can be associated with one another. Whether it is these ecological factors impacting the populations from recent changes or from generations prior,

changes must be made to ensure the benefit and cleanliness of our waterways are protected. This research provides the benefits of using a large database, Web of Science, on a diverse and populated invertebrate such as the *Hexagenia* mayfly to identify introductory materials on their lifespans but also the adaptations they have acquired through research already published.

LIMITATIONS

The limitations of this review include potential that determining factors were included in more than one search term category, such pollution and toxicity. This caused an inability to differentiate if there were other similarities within subcategories for both pollution and toxicity. Future efforts using more explicit search terms could to a more descriptive list stressor than what was identified during this review.

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