

Comparing detritus breakdown rates with and
without detritivores in subalpine ponds with
different hydroperiods

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Abstract

It is widely accepted that in stream systems, detritivores accelerate the breakdown of detritus that has been “conditioned” by microbial decomposers. By accelerating the breakdown of dead plant material, detritivores release nutrients and smaller food particles that are used by downstream plants and animals, respectively. Standing-water ecologists have often assumed animal detritivores play a similar role in ponds and wetlands, but there are few published studies investigating the contribution of animal detritivores to the overall breakdown of detritus in standing water systems. In this study, I compared detrital breakdown rates with and without invertebrate detritivores in nine subalpine ponds that differ in hydroperiod (permanent, semi-permanent, and temporary) at the Mexican Cut in the East River Valley in the Colorado Rockies. The biomass dominant detritivores in these systems are cased caddisflies (Trichoptera: Limnephilidae). I placed 14 containers with 5g of sedge into 3 permanent and 3 semi-permanent ponds, and 10 containers in 3 temporary ponds, into the nine ponds. half of the containers were covered with coarse mesh (20 mm) to allow detritivore access, while the other half were covered in fine mesh (2 mm) to prevent detritivore access. Every ten days, a pair of containers (one coarse and one fine mesh) were removed from each pond to compare the rate of detritus breakdown through time. . On each date, the sedge was oven-dried and weighed, and detritivores in each container were identified. Detritus in the coarse mesh containers lost mass two times faster than the detritus in the fine mesh containers. Fine particulate organic matter increased at a higher rate in the coarse mesh containers, suggesting that these caddisflies have a significant impact on the breakdown rate of detritus. Additionally, the detritus broke down fastest in the permanent ponds.

Introduction

In freshwater ecosystems, breakdown of dead organic plant material, or detritus, is the dominant energy pathway (Webster and Benfield 1986). Plant material is first decayed by the microbes (fungi and bacteria) that colonize the dead material. These microbes provide energy and nutrients for a host of macroinvertebrate detritivores by turning the unusable cellulose into usable carbon (Anderson et al, 1979, Webster et al, 1986, Hieber et al 2002, Chung et al, 2008). Detritivores are then the food source for predacious macroinvertebrates, such as dytiscid beetles, as well as salamanders, fish, and frogs (Cremona et al, 2018). A literature review by Webster and Benfield found that detritus breakdown rates are fastest in streams and slowest in wetland and pond systems, presumably because of differences in oxygen levels (Webster and Benfield 1986). This, however, only considered detrital breakdown in the absence of animal detritivores.

In stream systems, animal detritivores and microbial decomposers (fungi and bacteria) process and breakdown the majority of plant material that falls into the stream (Brinson et al. 1981). Many studies have shown that animal detritivores accelerate the breakdown of the organic matter in stream environments while feeding on the microbial decomposers that turn the detrital cellulose into digestible carbon (Anderson et al, 1979, Webster et al, 1986, Hieber et al 2002, Chung et al, 2008). Multiple studies have been conducted in stream systems that compare breakdown rates with and without animal detritivores; all have shown that in their presence, detritus breakdown rates are significantly higher (Cummins et al, 1973 and Herbst 1982). This “stream paradigm” for detritus breakdown emphasizes the role of animal detritivores in accelerating detritus breakdown as compared to microbial decomposition alone. However, direct evidence of the role animal detritivores has not been extensively studied in natural wetlands.

The stream paradigm may not apply to ponds and wetlands for several reasons. First, the proportion of detritus to detritivores is very low in many wetlands because of the extraordinary abundance

of aquatic plants (Batzer et al. 2015). Second, biofilms can collect more easily in wetlands and ponds due to the lack of running water, thus promoting a “microbial loop” in which algae and heterotrophic decomposers create a positive feedback that accelerates microbial breakdown (Keuhn et al. 2014). Additionally, since there is no flowing water in these systems, detritus does not experience the mechanical breakdown that it would in a stream system (Webster and Benfield 1986). Due to these factors, animal detritivores might make a trivial impact on detritus breakdown rates in many, if not all, wetland and pond systems. I know of only one published study that has compared the rate of detritus breakdown with and without detritivores under natural conditions in ponds (Wissinger et al. 2018), and three other unpublished studies (Wissinger, unpublished data). The published study found that detritivores increased detritus breakdown rates by 2-3 times that of microbes alone in montane kettle ponds near RMBL; however, the three unpublished studies found trivial effects of animal detritus on total breakdown rates in marshes and swamps in Pennsylvania (Wissinger and Bullis, unpublished data). Clearly, more studies are needed to determine what factors affect whether or not lentic detritivores make a measurable contribution to the overall rate of detritus breakdown, and hence to the mobilization of detrital energy and nutrients to wetland food webs (Balik et al. 2018).

The goal of this study was to determine the relative importance of microbial decomposers and animal detritivores to the overall breakdown of detritus in subalpine ponds at the Mexican Cut Nature Reserve. The only group of organisms that occur in these ponds that are described as shredders of detritus are caddisflies (Trichoptera: Limnephilidae) (see complete list of all species at Mexican Cut in Wissinger et al. 1999). I also wanted to compare breakdown rates in ponds with different hydroperiods, and hence difference densities and combinations of caddisfly detritivores. The species that dominates in permanent ponds (*Limnephilus externus*) processes detritus faster in microcosm experiments than the species (*Asynarchus nigriculus*) that dominates in temporary ponds (Wissinger et al. ms). Thus, I hypothesized that the role of animals detritivores would be greatest in permanent ponds, intermediate in semi-permanent

ponds (mix of the two species), and lowest in temporary habitats. I also predict that the amount of fine particulate organic matter (FPOM) will be positively related to the amount of coarse material that is processed in the containers.

Methods

Study Site

This project was conducted in nine subalpine ponds at the Mexican Cut in the East River Valley of the Colorado Rockies (table 1, figure 6). These ponds are all at an elevation ranging from 3,396 to 3,414 m. The dominant detritivores in these ponds are all Limnephilidae caddisflies, including *Asynarchus nigrificus* in the temporary ponds, *A. nigrificus*, *Limnephilus externus*, and *Limnephilus picturatus* in the semi-permanent ponds, and *L. externus*, *Agrypnia deflata*, and *Grammotaulius lorretae* in the permanent ponds. All containers were placed on the northeast side of each pond, at a depth of about 30 cm.

Experiment set-up

In order to measure detritus breakdown rates, 14 containers (17 x 15 x 10 cm) were placed into the permanent and semi-permanent ponds, and 10 containers in the temporary ponds. Half were covered with fine mesh (2 mm) to prevent detritivore access, while the other half were covered with coarse mesh (20 mm) to allow detritivore access. Each container was filled with 5g of air-dried sedge (*Carex aquatilis*)/grass mix. There were six 1 in² tiles and six glass marbles placed into each container for ballast. Before being placed into the ponds, the containers were filled with approximately a liter of pond water and set pondside for 72 hours to saturate the sedge, preventing the detritus from floating out of the containers when submerged. After the 72 hour saturation period, six containers (three fine mesh and three coarse mesh) were brought back to the lab, oven dried at 60°C for 48 hours, and weighed, in order to assess the mass lost due to leaching. There was an additional pair of empty containers (one fine mesh and one coarse mesh) placed into each pond without sedge as a control for the accumulation of any material that may have fallen into the other containers throughout their time in the ponds.

Data collection

The containers were randomly selected for removal without replacement every 10 days. Containers were placed immediately into buckets after removal from the pond in order to prevent loss of material. The contents of the containers were placed into a sorting tray in order to remove the animals. All animals were removed from the trays, identified to species, and placed back into their original pond. The detritus was then filtered through a 1 mm net to separate coarse particulate organic matter (CPOM) from the fine particulate organic matter (FPOM). They were brought back to the lab and oven dried for 48 hours at 60°C, then weighed.

Data analysis

In order to find the starting mass of the sedge within the containers when they were placed into the ponds, the average of mass lost between air drying and oven drying, as well as the average mass lost during leaching, was subtracted from the initial sedge weights. To account for any material that may have fallen into the containers during their time in the pond, the mass of the organic matter that fell into the empty control containers was subtracted from the remaining coarse or fine containers. To quantify the breakdown rates of detritus, the mass lost was calculated through time for each pond, and; the rate of conversion from CPOM to FPOM was also calculated.

Results

By the end of 6 weeks, the detritus in the coarse mesh containers broke down 5 times faster than the detritus in the fine mesh containers ($p < 0.0001$) (fig 1). The faster breakdown of the detritus in the fine mesh containers during the first 18 days might be attributed to leaching of dissolved organic compounds that overshadowed the breakdown rate of the microbes, which is most likely the rate from days 19-39. In the coarse meshed containers, detritus broke down faster in the permanent than in the semi-permanent ponds ($p < 0.0001$) (fig 2). The detritus in the fine mesh containers broke down at an rate across the two pond types (fig 2). There is a weak relationship ($R^2 = 0.187$) between these breakdown rates and the

density of caddisflies in each pond type (fig 3). When looking at the densities of *Limnephilus externus*, the most abundant caddisfly shredder in each pond, the relationship becomes slightly stronger ($R^2 = 0.247$); however, it is still weak overall (fig 4). It was also shown that as more FPOM accumulated, there was less remaining CPOM (fig 5). I could not compare the breakdown rates in the temporary ponds to the other two pond types because of the extraordinarily early drying of those ponds.

Discussion

Although the importance of detritivores in stream systems is well-known, there are few published studies that examine their role in pond and wetland systems with regards to detritus breakdown rates. These results indicate that in these pond systems, macroinvertebrate detritivores, especially limnephilid caddisflies, have significant impacts on the breakdown rates of detritus. I found that detritus breaks down at a faster rate in the presence of detritivores, as compared to microbes alone. These results correspond to a previous study in montane ponds near RMBL where caddisflies were increased detritus 2-3 fold as compared to microbes only. (Wissinger et al 2018). The results from these two studies show that caddisflies, specifically of the Limnephilidae family, have significant impacts on the breakdown rates of detritus in ponds.

The results of this study are also consistent with the many stream system studies that have focused on effects of detritivores on breakdown rates of detritus. A 1982 study performed in two streams in North Carolina demonstrated that plant material in the stream where invertebrates were removed decayed much slower than in the stream with macroinvertebrates (Wallace et al, 1982). It was also seen that there was less FPOM created; therefore there was not as much food produced for animals downstream (Wallace et al, 1982). I also found that the containers where detritivores have access to the detritus, there was significantly more FPOM produced. This fine particulate organic matter is important in stream systems because it flows downstream as food for other organisms (Wallace et al, 1982). It is not yet known the role that FPOM

plays in the wetland and ponds, but we do know that more of it is produced in the presence of detritivores. In a similar microcosm experiment, it was found that detritus breakdown was much faster when macroinvertebrates were present (Martinez et al 2014). This study also used microcosms, or small containers, to observe the breakdown rates of detritus in headwater streams.

We may have seen such rapid breakdown rates because we conducted this experiment from June-August 2018, which corresponds to the the warmest water temperatures and highest densities of caddisflies in these ponds (Wissinger et al. 2003). . It is known that warmer water temperatures lead to faster rates of microbial breakdown (Brock 1984, Martinez et al 2014). Because of this, we are leaving a pair of containers in each pond during the winter to be harvested the summer of 2019 to obtain a total annual breakdown rate, and hence the actual contribution of microbes and detritivores on an annual basis.

There have been three unpublished studies performed in Pennsylvania that all used very similar methods to this experiment. Each had containers filled with detritus placed into ponds. Some containers allowed detritivore access, while the others prevented it. However, in all three of those studies it was found that detritivores had trivial effects on the breakdown rates of detritus. One reason we may be seeing differences in results due to the varying inputs of detritus in each of these systems. In the ponds in Pennsylvania, there is a significantly larger detrital input compared to the ponds at the Mexican Cut. In order to compare these studies to each other, we need to determine the yearly inputs of detritus into each system. By analyzing this, we will be able to observe if this truly does make an impact on breakdown rates of detritus. Additionally, the ponds at the Mexican Cut have a significantly higher density of caddisflies compared to the ponds in northwestern Pennsylvania. This variation in caddisfly density may also play a role in why we are getting different results in different systems, all while using the same methods. In order to investigate why we are getting different results while using similar methods in similar environments, I plan to compile and create a comparative data set from the multiple studies conducted across different pond types. Ideally, I will be able to identify the variables, such as density of shredders, annual detritus

input, detritus type, and pond hydroperiod, that affect whether detritivores have a trivial vs. large effect on overall breakdown rates in shallow standing waters.

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Tables and figures

Pond	Elevation (m)	Longitude	Latitude
1	3410	-107.0652231	39.02916932
5	3404	-107.0632858	39.02773147
6	3405	-107.0633859	39.02815677
7	3405	-107.063614	39.02840328
8	3406	-107.0639098	39.02902008
10	3406	-107.064297	39.02974849
12	3408	-107.0648848	39.0295607
13	3414	-107.0642476	39.02811601
15	3396	-107.0625382	39.0281756

Table 1.) Elevation and GPS coordinates of all nine ponds.

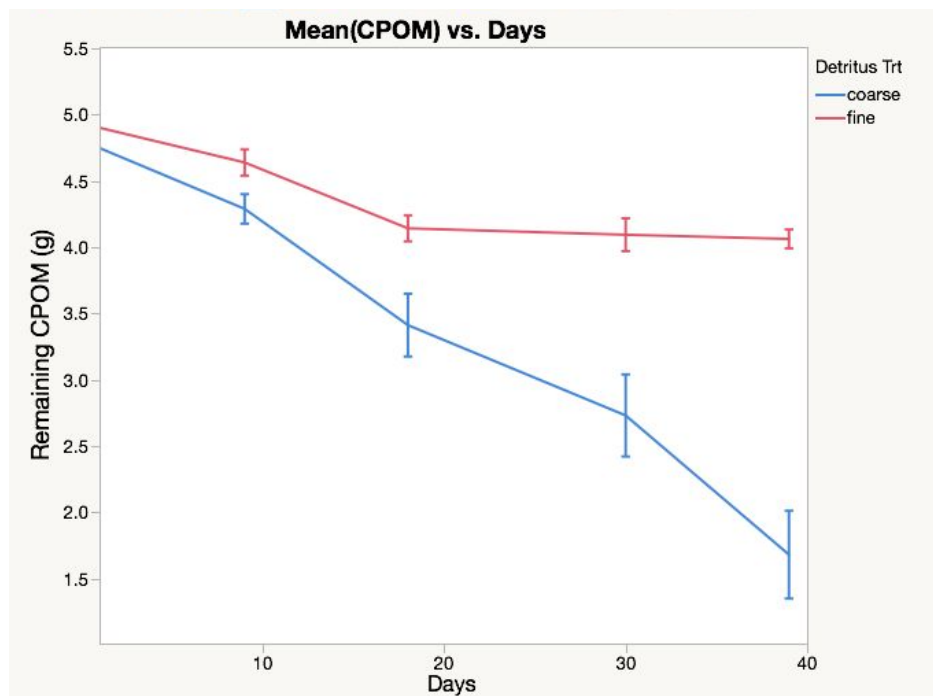


Figure 1.) Mass of remaining CPOM in the fine and coarse mesh containers. The detritus in the coarse mesh containers (blue line) lost mass faster than the detritus in the fine mesh (red line) containers. ($p < 0.0001$)

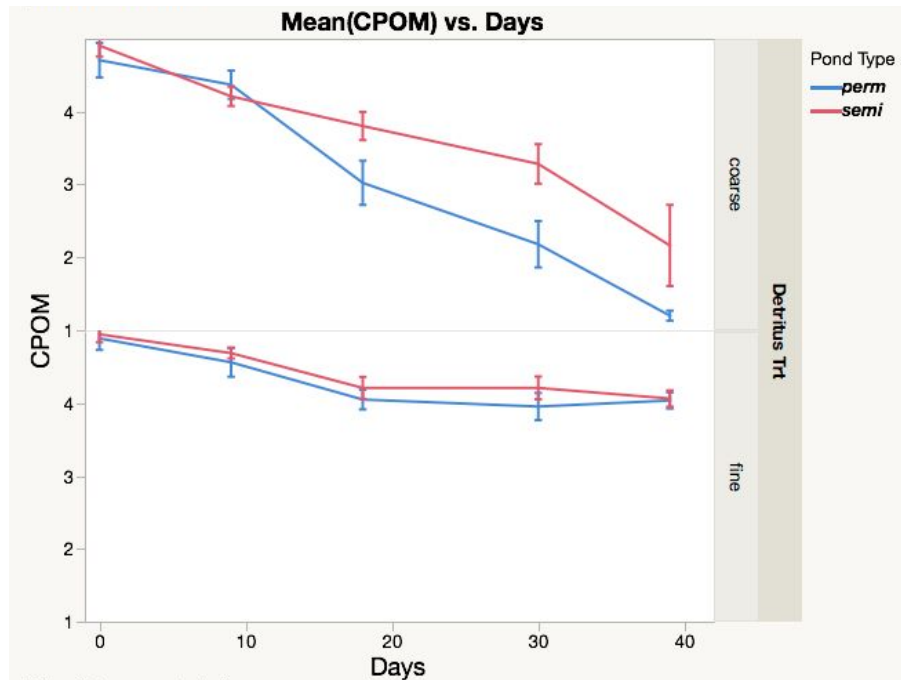


Figure 2.) Mass of CPOM remaining in semi-permanent vs permanent ponds. The fine mesh containers had little variation. Detritus in the coarse mesh containers broke down faster in permanent ponds compared to semi-permanent ($p < 0.0001$)

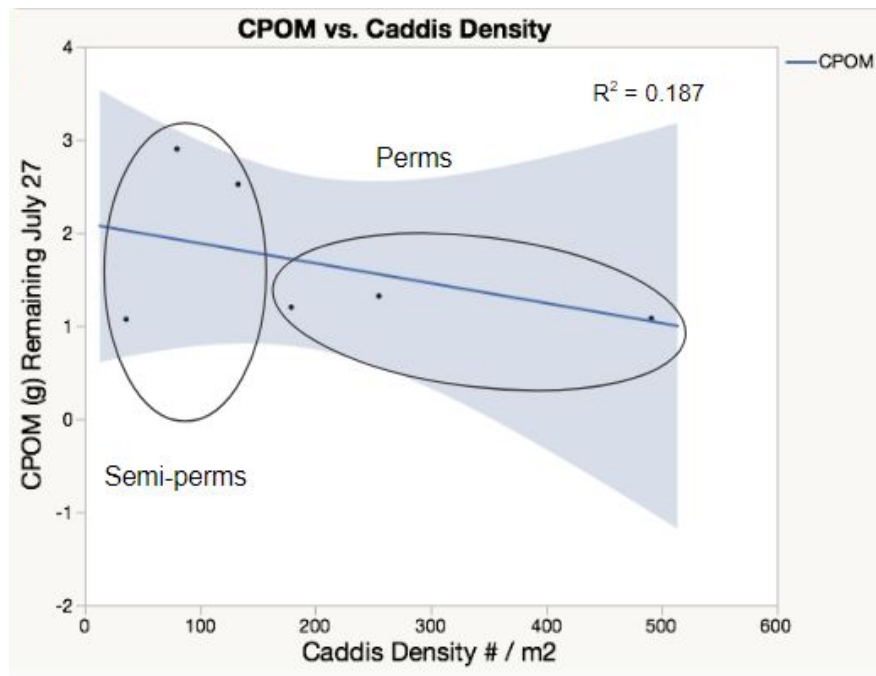


Figure 3.) Grams of CPOM remaining after 40 days in the pond vs density of total caddisflies in the pond. A weak relationship can be seen, even though the three permanent ponds have higher densities of caddisflies ($R^2 = 0.187$).

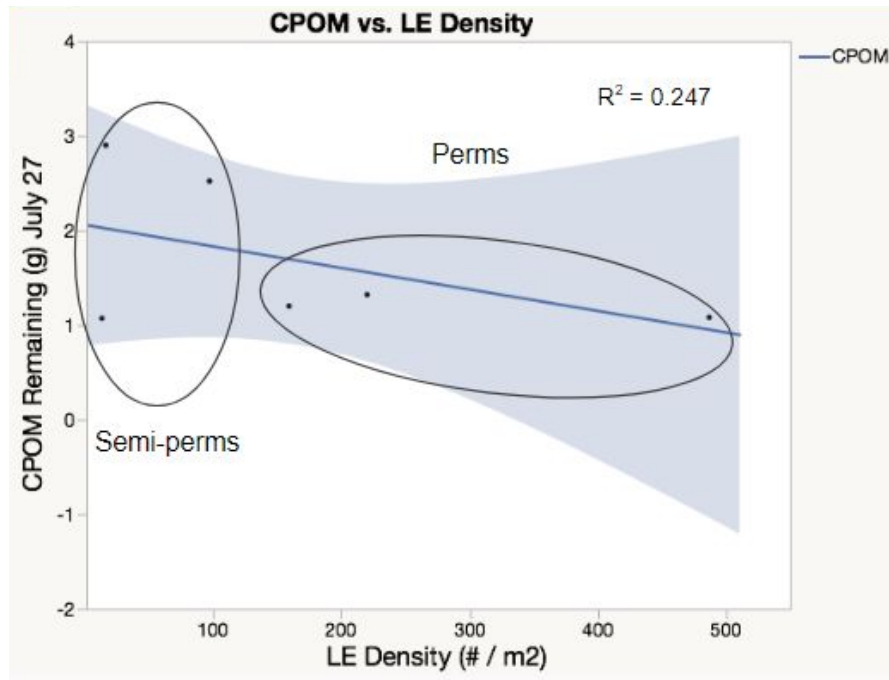


Figure 4.) Grams of CPOM remaining after 40 days in the pond vs the density of *Limnephilus externus* caddisflies. A weak relationship can be seen, even though the three permanent ponds have higher densities of caddisflies ($R^2 = 0.247$)

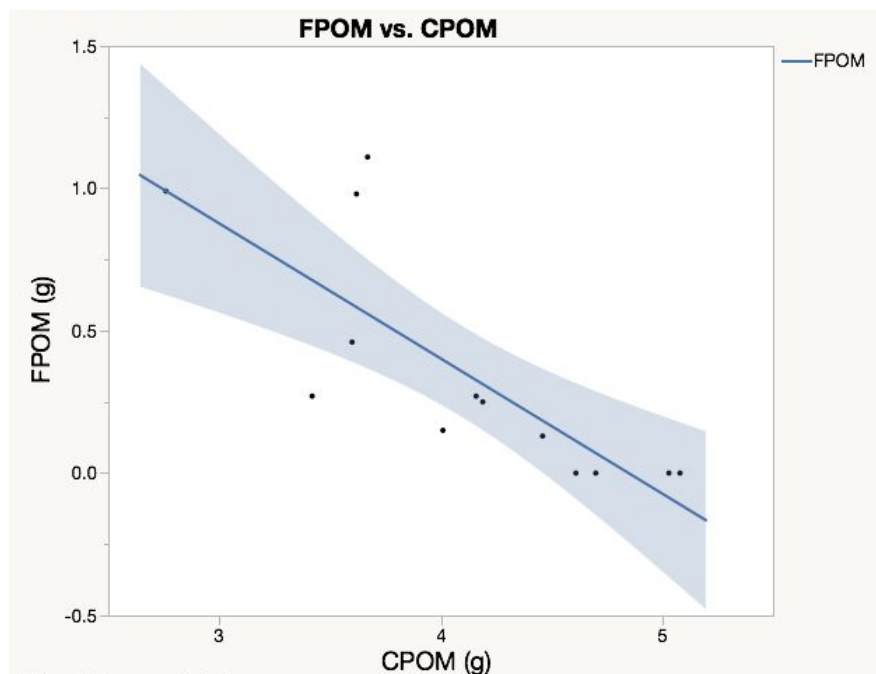


Figure 5.) The amount of FPOM in the containers as a function of CPOM remaining.

2018 Mexican Cut Pond Sites

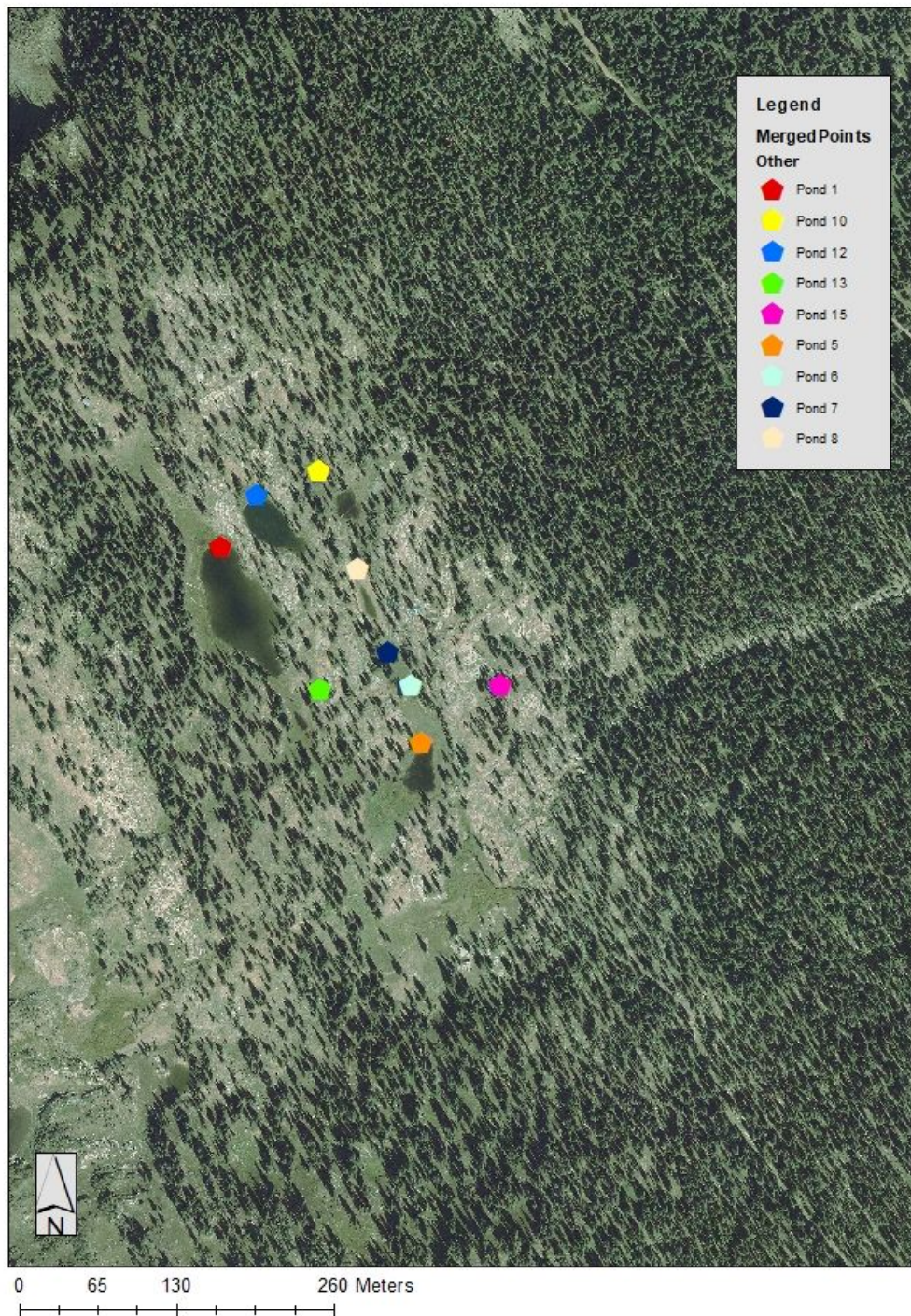


Figure 6.) GIS map of all nine pond sites at the Mexican Cut.

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