



Investigation of Optical Brightening Agents (OBAs) in the Major Lakes of the Clearwater Watershed in Montana

Clearwater Resource Council

Seeley Lake, Montana

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EXECUTIVE SUMMARY

Optical brightening agents (OBAs) are chemical compounds that are added to products such as laundry soaps, detergents, or cleaning agents. The presence of OBAs in water systems to which wastewater is being discharged can indicate failing septic systems or a lack of proper water treatment. This study, which sampled and tested surface water for the presence of OBAs, was designed to help identify the potential septic contamination to six lakes in the Clearwater Watershed. After collecting samples and completing fluorometric testing, all of the samples were concluded to be negative for optical brightening agents. However, it should not be concluded that OBAs are not present in the lakes that were studied. This study had a few limitations that suggest the results may underestimate the presence of OBAs. Further study is needed to fully understand the presence of OBAs and septic leachate in the Clearwater Watershed.

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1.0 ACKNOWLEDGMENTS

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- Tom Joehler: Placid Lake
- Joann Wallenburn: Seeley Lake
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- Flathead Lake Biological Station
- Missoula County Conservation District
- Montana Department of Natural Resources

2.0 INTRODUCTION

2.1 Purpose

Optical brightening agents (OBAs), also sometimes referred to as optical brighteners (OBs), are chemical compounds that are added to products such as laundry soaps, detergents, or cleaning agents. These compounds adsorb to fabrics during the laundering process for the purpose of making fabrics appear brighter. OBAs can be found in essentially all laundry detergents.

Laundry wastewater retains a large portion of dissolved optical brightening agents even after the laundering process is complete. The wastewater from laundry is typically discharged through a household's sewer or septic system. In the Clearwater Watershed, nearly every household and business is connected to an individual or clustered septic system. As seen in Figure 1, septic systems generally have two main parts. First, wastewater is piped into the septic tank. This enclosure is designed to ensure the solid wastes sink to the bottom of the tank. Then, the remaining liquid flows out to the drainfield for final treatment. The drainfield utilizes the natural properties of soil to filter and clean the liquid. Ideally, fully treated and neutralized liquid makes its way through the septic system, to the ground water, and eventually into the nearby surface waters such as lakes, rivers, and streams. OBAs are highly susceptible to adsorption in soil and therefore are removed from wastewater in a well-functioning septic system. However, septic systems, particularly those that are old and/or not well maintained, can fail, thus introducing a variety of undesired pollutants and OBAs into the ground water and surface water.

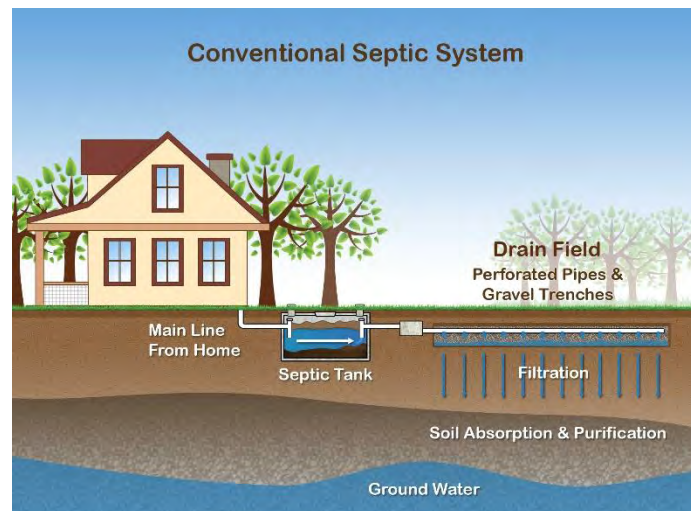


Figure 1: Septic System Diagram (Minnesota Pollution Control Agency)

The presence of OBAs in water systems to which wastewater is being discharged can indicate failing septic systems or a lack of proper water treatment. Moreover, OBAs serve as ideal indicators of failing septic systems because they can be detected and measured by a device called a fluorometer. This study, which sampled and tested surface water for the presence of OBAs, was designed to help identify the potential septic contamination to six lakes in the Clearwater Watershed.

2.2 Study Area

The Clearwater watershed (Figure 2), located in Missoula County, Montana, has unique natural resource values as it forms the southernmost portion of the North Continental Divide Ecosystem (NCDE), which extends from the Highwood River in Alberta to the Blackfoot River in Montana. The NCDE is also known as the “Crown of the Continent” and encompasses over 10 million acres of some of the most unspoiled lands on the entire North American continent. Glacial forces shaped the Clearwater valley’s unique topography over two million years ago, and formed the succession of lakes, which from north to south include: Rainy Lake, Lake Alva, Lake Inez, Seeley Lake, and Salmon Lake. The economic importance of this chain of lakes in addition to nearby lakes such as Big Sky Lake and Placid Lake is undeniable since the hub of activity here in the Clearwater occurs in close proximity to these bodies of water. They provide prime recreational opportunities, such as fishing, swimming, and boating, for both residents and tourists.

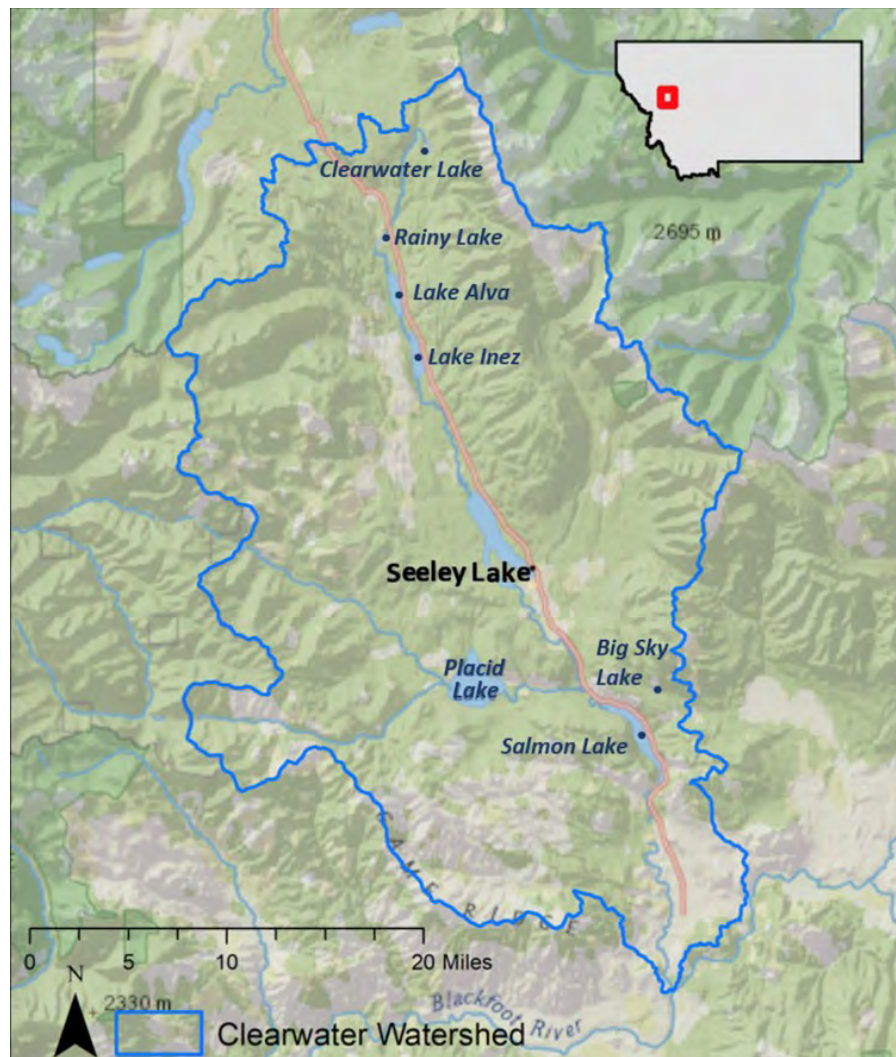


Figure 2: Map of Clearwater Watershed

Lake Alva, Lake Inez, Seeley Lake, Salmon Lake, Big Sky Lake, and Placid Lake are the major lakes in the Clearwater Watershed and were selected to be sampled for this study. With the exception of Lake Alva, all of these lakes have been residentially developed to varying extents. The maps in Appendix A show the location of septic systems on each of the lakes.

3.0 METHODOLOGY

3.1 Sampling Frequency, Locations, and Techniques

The six major lakes in the Clearwater Watershed were sampled throughout the summer of 2022 beginning in July and ending in September. There were five rounds of sampling on Seeley Lake throughout the summer and four rounds of sampling on the other lakes. Table 1 lists the sampling dates for all the lakes.

Sampling Dates					
Lake	Sampling #1	Sampling #2	Sampling #3	Sampling #4	Sampling # 5
Seeley	7/11/2022	8/1/2022	8/15/2022	9/6/2022	9/12/2022
Placid	7/14/2022	8/5/2022	8/19/2022	9/9/2022	
Alva	7/19/2022	8/2/2022	8/16/2022	8/30/2022	
Inez	7/20/2022	8/3/2022	8/17/2022	9/17/2022	
Salmon	7/21/2022	8/4/2022	8/18/2022	9/1/2022	
Big Sky	8/8/2022	8/22/2022	8/29/2022	9/12/2022	

Table 1: Sampling dates for each lake

A varied amount of sample sites were chosen for each lake based on lake size and population density. Seeley Lake has ten sites, Placid Lake has four sites, Lake Alva has one site, Lake Inez has four sites, Salmon Lake has five sites, and Big Sky Lake has one site. Table 2 below details the ID and location of each site. Every lake has one site that corresponds to the deepest point in the lake. Most of the other sites are located near the lake shore. Maps depicting the location of all the sample sites can be found in Appendix B.

The sites chosen for the OBA sampling are the same sites used in Clearwater Resource Council’s regular aquatics sampling. This overlap in sites allows for potential further data analysis and contextualization.

Lake	Site ID	Description	Latitude	Longitude
Big Sky	BS1	Deep (50 ft / 15.2 m)	47.116	-113.396
Alva	AL1	Deep (90 ft / 27.4 m)	47.316	-113.583
Inez	IZ4	Surface	47.271	-113.569
	IZ3	Surface	47.278	-113.561
	IZ1	Surface	47.287	-113.571
	IZ2	Deep (74 ft / 22.6 m)	47.279	-113.565
Placid	PL4	Surface	47.108	-113.519
	PL3	Surface	47.133	-113.529
	PL2	Surface	47.119	-113.504
	PL1	Deep (93 ft / 28.3 m)	47.118	-113.521
Salmon	SA3	Surface	47.076	-113.388
	SA1	Surface	47.069	-113.386
	SA4	Surface	47.1	-113.403
	SA5	Surface	47.105	-113.41
	SA2	Deep (60 ft / 18.3 m)	47.097	-113.405
Seeley	SE3	Surface	47.174	-113.482
	SE4	Surface	47.173	-113.492
	SE5	Surface	47.193	-113.508
	SE7	Surface	47.204	-113.512
	SE8	Surface	47.213	-113.522
	SE6	Surface	47.19	-113.515
	SE1	Deep (60 ft / 18.3 m)	47.175	-113.489
	SE2	Surface	47.186	-113.504

Table 2: Sample sites and locations

For sites describes as “surface” in Table 2 above, “grab” samples were taken in a clean 250 ml Nalgene bottle. Immediately prior to taking the sample, the bottle was rinsed out three times with sample site water. The grab sample was taken by dunking and filling the Nalgene bottle under the water at about forearm depth. For sites described as “deep” in Table 2 above, a Van Dorn style self-closing sampler was used. Immediately prior to taking the sample, both the Van Dorn sampler and a 250 ml Nalgene bottle were rinsed out three times with sample site water. Then, the Van Dorn sampler was lowered down to collect sample water from one meter above the lake’s bottom. This sample water was transferred to the 250 ml Nalgene bottle. Both surface and deep samples were immediately covered with aluminum foil to prevent photo-decay of the OBAs.

3.2 Sample Analysis Procedures

The Turner Designs *Aquaflor* handheld fluorometer was used to detect and quantify the amount of OBAs in the collected samples. OBAs are excited by wavelengths of light in the near-ultraviolet (UV) range (360 to 365 nm) and then emit light in the blue range (400 to 440 nm). Electrons in fluorescent molecules are excited into a higher energy state by absorption of light and then emit a small amount of heat plus fluorescence as the electrons return to their ground state (Burres, 2011). The fluorescence from the second excited state is measured by the fluorometer.

Fluorometric calibration was conducted using a 50ppm OBA calibration solution made from Tide detergent and DI water. The calibration solution was made with instructions from California State Water Resources Control Board's Standard Operating Procedure 3.4.1.4 for "Measuring Optic Brighteners in Ambient Water Samples Using a Fluorometer" (Burres, 2011).

After calibration was complete, the fluorometer was ready to test samples. Triplicates were analyzed for each sample. The fluorometer provides results in parts per million (ppm) which is equivalent to relative fluorescence units (RFU). The following steps describe the sample analysis procedure as detailed in SOP 3.4.1.4:

Step 1

Measure initial fluorescence using *Aquaflor*.

- If the sample measures <5ppm, conclude that the sample is negative for optical brighteners.
- If the sample measures >5ppm, continue to step 2.

Step 2

Expose samples directly to UV light for 5 minutes and then measure fluorescence again. Calculate the percentage of reduction in fluorescence after 5 minutes compared to before UV exposure.

- If % reduction <8%, conclude the sample is negative for optical brighteners.
- If % reduction $\geq 30\%$, conclude the sample is positive for optical brighteners.
- If % reduction <30% and >8%, continue to step 3.

Step 3

Expose samples under UV for another 5 minutes, measure fluorescence, calculate the ratio of % reduction in fluorescence after 10 minutes of cumulative UV exposure over % reduction after 5 minutes UV exposure.

- If the ratio is equal ≥ 1.5 , conclude that the sample is negative for optical brighteners.
- If the ratio <1.5, conclude that the sample is positive for optical brighteners.

Step 4

Out of the three replicates:

- If all three are positive, conclude that the sample is positive for optical brighteners.
- If two out of three are positive, conclude that the presence of optical brighteners within the sample is undetermined.

For the purpose of analysis, the initial fluorescence of the triplicates was averaged. Figure 3 below shows the analytical procedure in diagram form.

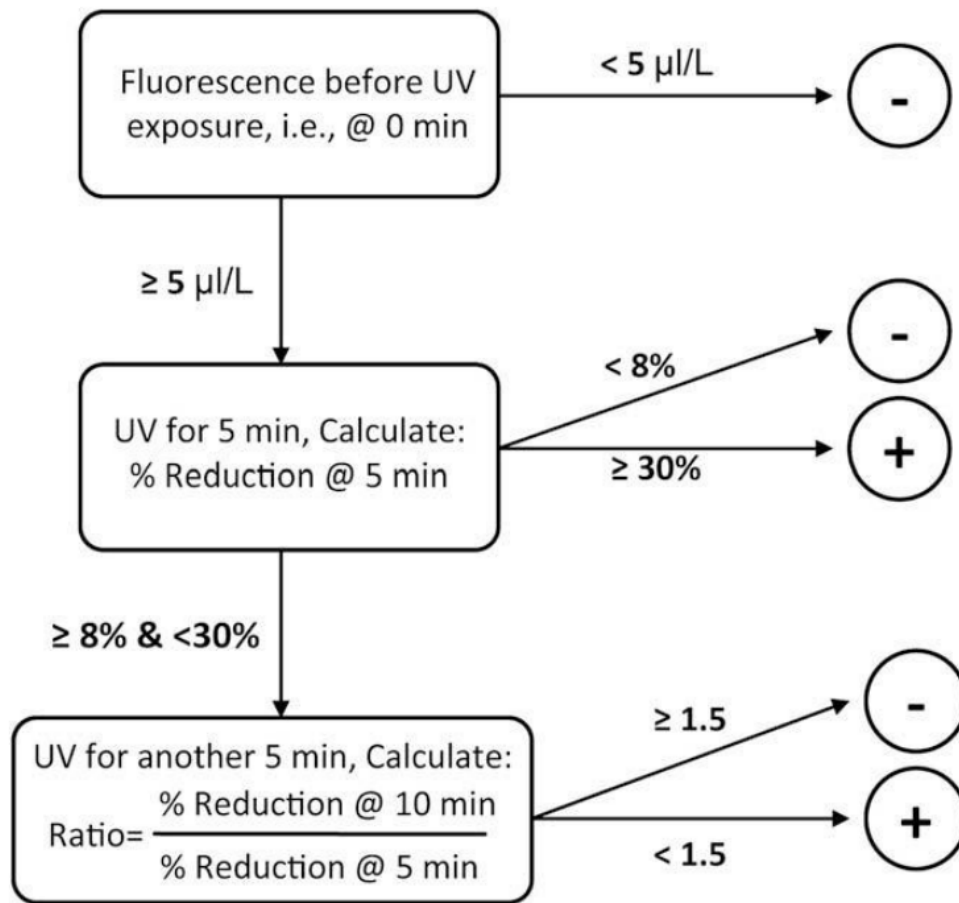


Figure 3: Diagram of Analytical Procedure (Burres, 2011)

4.0 RESULTS

A total of 90 samples were collected. However, two of the samples were unable to be analyzed. The sample from site SE14 on 8/15/22 drained during storage prior to testing. The sample from site IZ3 on 8/17/22 was exposed to light for an extended period of time during storage. This exposure to light resulted in photo-decay of the potential OBAs in the sample, rendering the sample untestable. In result, there were a total of 88 samples tested. Appendix C contains the full data sheet for the fluorometer testing.

The range of average initial fluorescence was about 3.59 RFU. The maximum average initial fluorescence value of 4.696 RFU occurred on 8/15/22 at site SE2. The minimum average initial fluorescence value of 1.099 RFU occurred on 9/1/22 at site SA4.

Only three of the initial fluorescence value tests resulted in a RFU value greater than 5. Those samples were IZ2 on 8/3/22 (first triplicate tested), PL2 on 8/19/22 (first triplicate tested), and SE2 on 9/20/22 (first triplicate tested). However, the percent reduction calculated in round two concluded those samples were negative for OBAs. Moreover, the other two triplicates for those samples tested below 5 RFU at 0 minutes. Every other sample tested below 5 RFU at 0 minutes. Therefore, all the samples were concluded to be negative for OBAs. The average initial fluorescence values for all 88 samples are listed in the tables below.

Seeley Lake Average Initial Fluorescence Values (RFU)										
Date	SE2	SE4	SE5	SE8	SE9	SE10	SE11	SE12	SE13	SE14
7/11/2022	3.516	2.266	2.196	1.658	2.444	2.672	2.658	3.632	2.692	2.048
8/1/2022	3.747	2.055	2.264	2.223	1.816	1.935	2.427	2.270	2.604	2.738
8/15/2022	4.696	1.500	2.923	2.135	1.853	1.803	2.328	3.257	2.512	
9/6/2022	3.879	1.815	1.967	1.742	1.805	2.017	1.848	1.376	2.082	2.255
9/12/2022	3.875	1.496	1.922	2.742	2.710	2.535	2.522	2.250	2.201	3.510

Table 3: Seeley Lake Average Initial Fluorescence Values

Placid Lake Average Initial Fluorescence Values (RFU)				
Date	PL1	PL2	PL3	PL4
7/14/2022	3.048	1.825	2.219	1.896
8/5/2022	4.084	1.777	1.651	2.329
8/19/2022	2.305	3.802	2.733	1.587
9/9/2022	3.499	2.203	1.862	1.803

Table 4: Placid Lake Average Initial Fluorescence Values

Salmon Lake Average Initial Fluorescence Values (RFU)					
Date	SA1	SA2	SA3	SA4	SA5
7/21/2022	1.645	3.251	2.823	2.442	1.897
8/4/2022	2.544	3.975	1.828	1.460	2.152
8/18/2022	2.275	2.331	2.026	2.846	1.759
9/1/2022	1.888	2.930	1.482	1.099	2.048

Table 5: Salmon Lake Average Initial Fluorescence Values

Lake Inez Average Initial Fluorescence Values (RFU)				
Date	IZ1	IZ2	IZ3	IZ4
7/20/2022	1.980	3.150	1.858	1.884
8/3/2022	1.731	3.950	2.331	2.458
8/17/2022	2.530	2.042		1.829
9/17/2022	1.458	2.841	2.040	2.391

Table 6: Lake Inez Average Initial Fluorescence Values

Lake Alva Average Initial Fluorescence Values (RFU)	
Date	AL1
7/19/2022	3.745
8/2/2022	3.689
8/16/2022	3.362
8/30/2022	3.219

Table 7: Lake Alva Average Initial Fluorescence Values

Big Sky Lake Average Initial Fluorescence Values (RFU)	
Date	BS1
8/8/2022	2.819
8/22/2022	3.007
8/29/2022	2.803
9/12/2022	2.431

Table 8: Big Sky Lake Average Initial Fluorescence Values

5.0 DISCUSSION

5.1 Analysis of Results

All the samples tested were concluded to be negative for optical brightening agents. The average variance for all sites across the sampling season is 0.212. On the other hand, the average variance for all sampling dates across the sampling sites is 0.442. The fluorometer values varied more spatially across different sites than they varied temporally across different sampling dates. In other words, there was not a clear trend in fluorometric values throughout the sampling season. The data does suggest, however, the fluorometric results remain relatively stable at each site throughout the sampling season.

5.1 Limitations of Study

There are four main limitations that have a potential impact on the outcome of this study. First, due to the timing of funding, sampling for this study did not begin until mid-July. July is notably after the end of the spring runoff period. Given that the spring runoff has an impact on ground water, surface water, and potentially septic systems, fluorometric testing during this time period would have been ideal. Second, except for the six mid-lake deep sites, all of the samples were collected via “grab” samples. Grab samples collect water from the top of the water column. However, groundwater has been shown in numerous studies to transport septic leachate through lake-bottom sediments into lake waters (Kerfoot and Brainard 197787; Belanger et al. 1985; Jourdonnais et al. 1986). Third, septic leachate plumes are temporally and spatially inconsistent. As described in the 2012 Whitefish Lake Institute’s report titled *Investigation of Septic Leachate to the Shoreline Area of Whitefish Lake, Montana*, “because fluorescence decreases over time with exposure to sunlight (Jourdonnais and Stanford, 1985), the best determination of septic leachate through fluorometry would occur in an area where leachate inputs are continuous. Given the sporadic nature of home inhabitation around the lake, septic system usage—and therefore wastewater movement and septic leachate inputs—are inconsistent” (Whitefish Lake Institute, 2012). The combination of inconsistent septic leachate plumes and the effects of photo-oxidation on OBAs suggests that the study results may underestimate the presence of septic leachate in the six lakes studied. Finally, studies show that the fluorometric testing method used can be “insensitive, in that it failed to detect raw sewage and approximately one-half of the detergents tested in natural stream water at 5 µl/L” (Cao et al. 2009).

6.0 CONCLUSION & RECOMMENDATIONS

The Clearwater Valley has a unique set of lakes that serve a variety of beneficial values. The economic importance of all six lakes is undeniable. They are the hub of recreational opportunities including fishing, swimming, and boating. Seeley Lake, in particular, is the center

of economic activity for a town of over 1,600 people. The six lakes in this study also serve important cultural and environmental functions. The watershed has unique cultural values to both the Confederated Salish and Kootenai Tribes and the Blackfoot Nation who referred to this area as the “backbone of the world.” The landscape and the species that reside on it rely on the watershed to maintain balance and health. From the endangered bull trout and iconic grizzly bears to the residents of Seeley Lake, protecting the water for generations to come is of paramount importance.

This study sampled and tested six lakes in the Clearwater Watershed for the presence of optical brightening agents, a group of chemical compounds that are indicators of potential septic leachate. While all the samples in this study were concluded to be negative for OBAs, it should not be concluded that OBAs are not present in the lakes that were studied. This study had a few limitations that suggest the results may underestimate the presence of septic leachate. Further study is needed to fully understand the presence of OBAs in the Clearwater Watershed.

Recommendations for a future study are as follows:

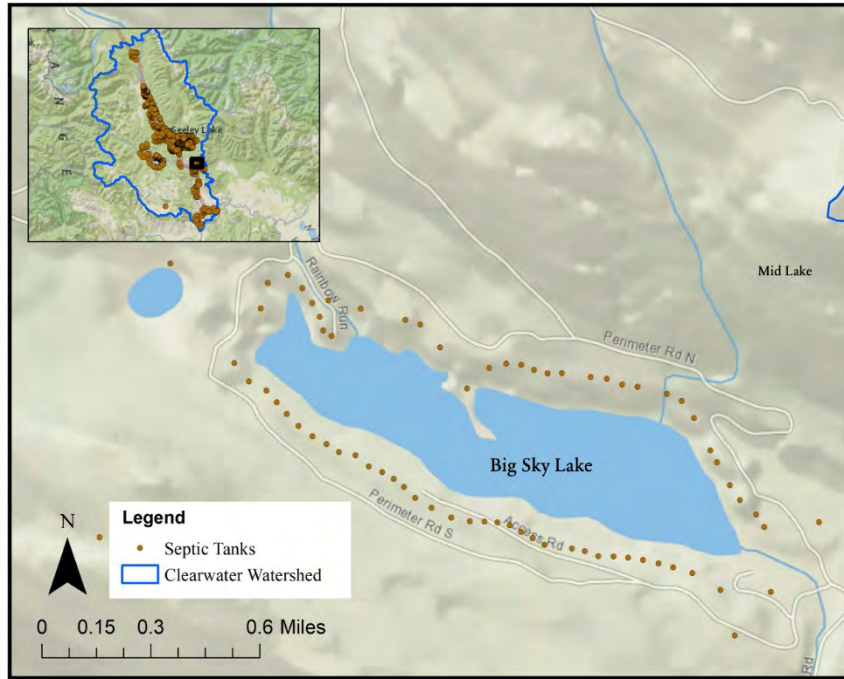
1. Begin the first round of sampling during spring runoff.
2. Utilize a Van Dorn style sampling device to collect samples no more than 1 foot from the bottom of the lake at each site.

As a measure to protect the Clearwater Watershed from potential degradation due to septic inputs while studies are ongoing, we recommend the continuation of community education and outreach programs. Property owners can be educated about septic system maintenance and the consequences of poorly maintained or updated systems.

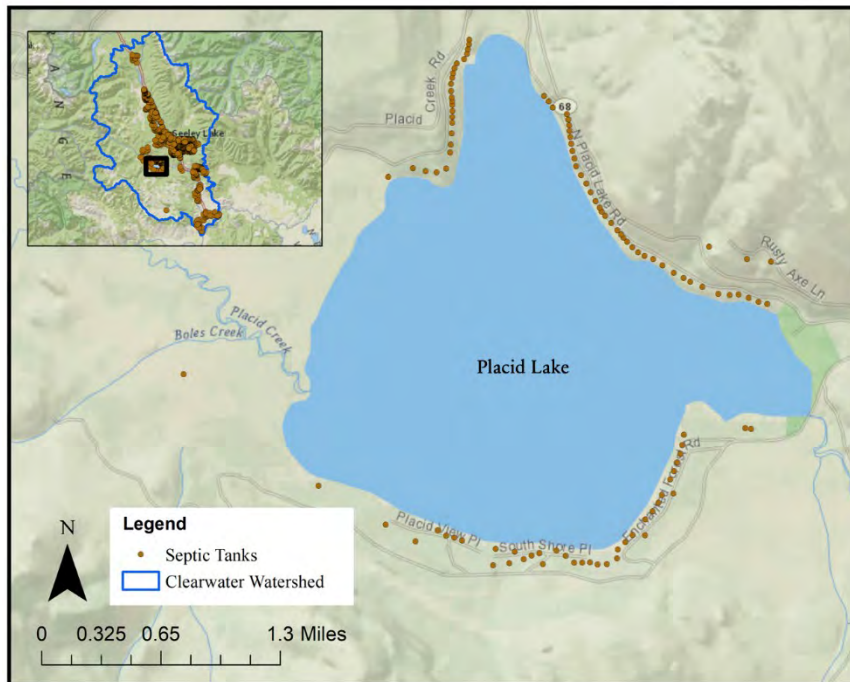
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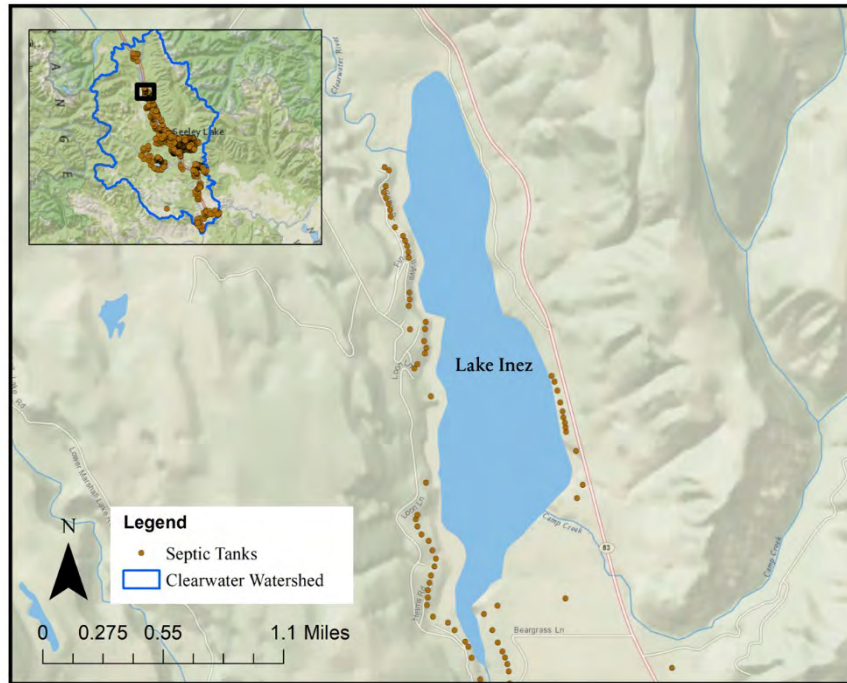
APPENDIX A: Septic System Densities



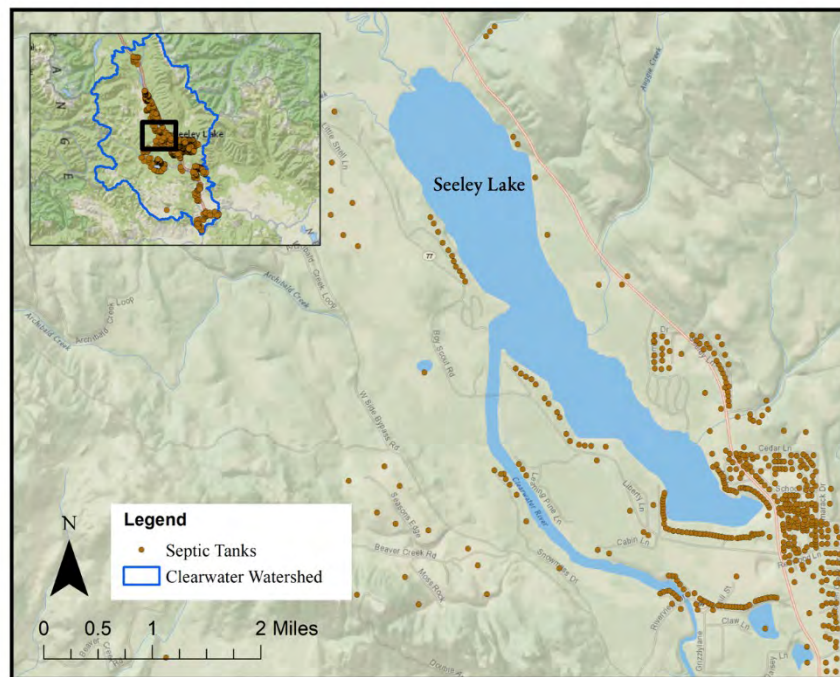
Big Sky Lake



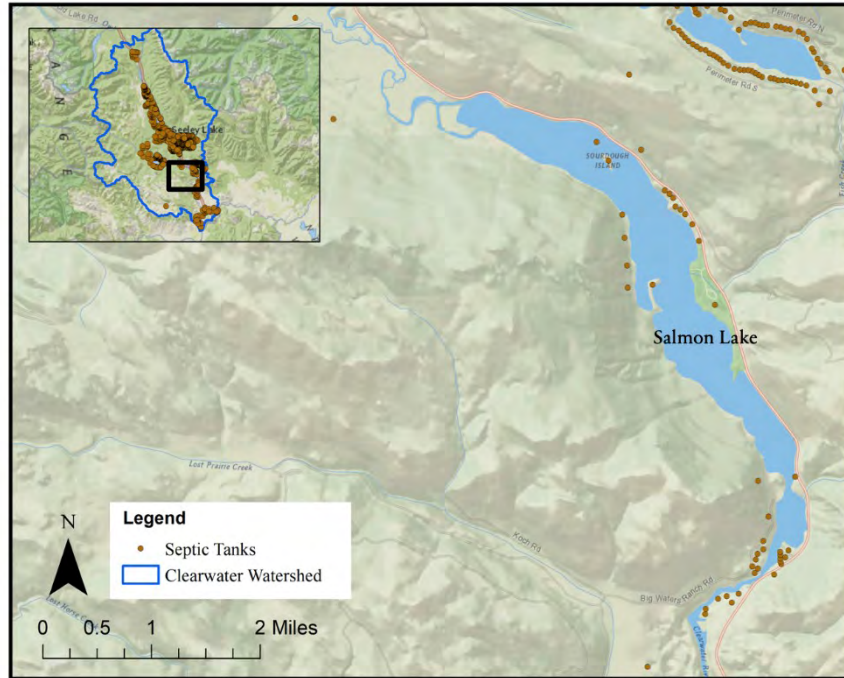
Placid Lake



Lake Inez



Seeley Lake



Salmon Lake

Note: There are no septic systems on Lake Alva.

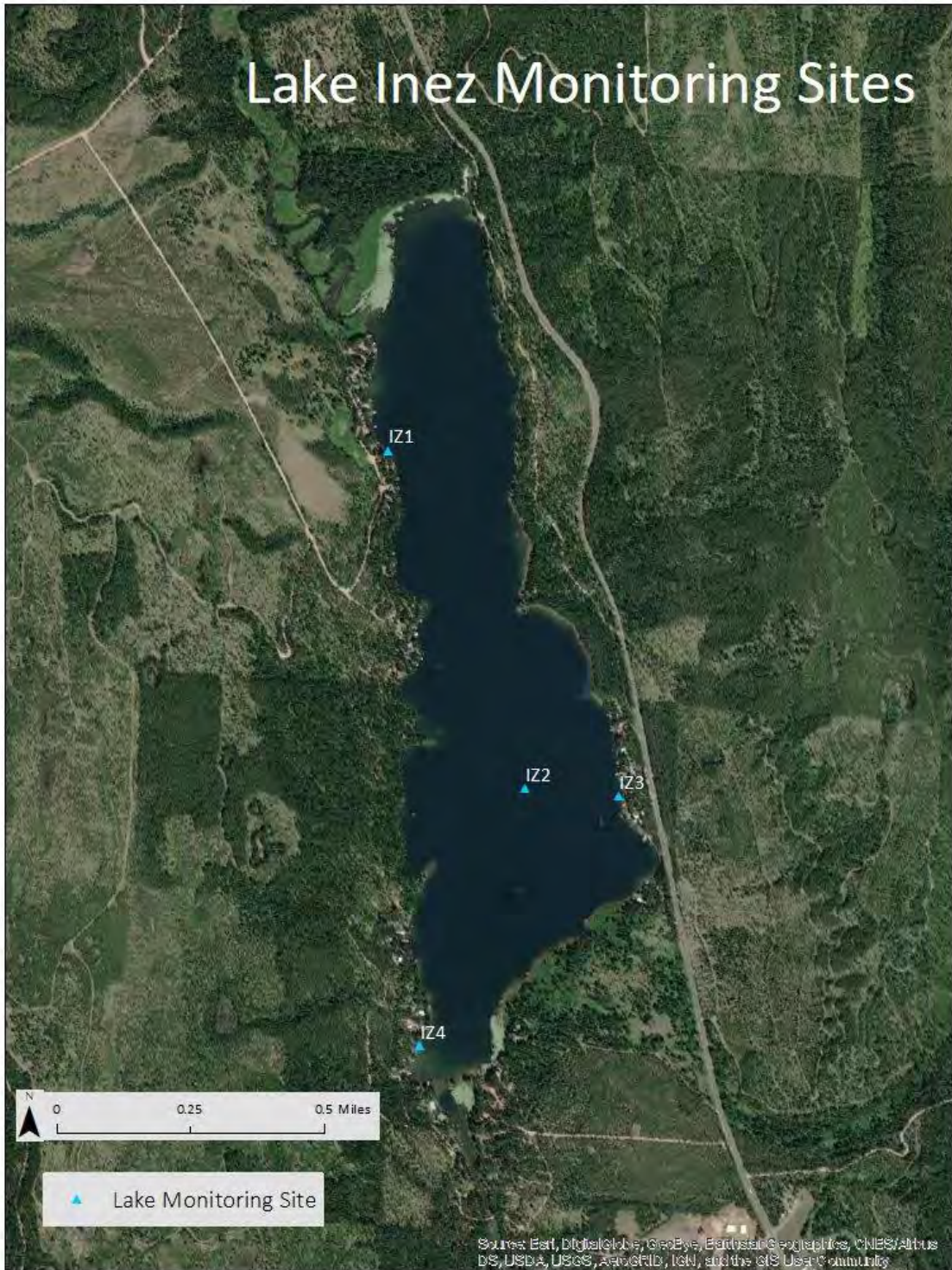
APPENDIX B: Sample Site Maps



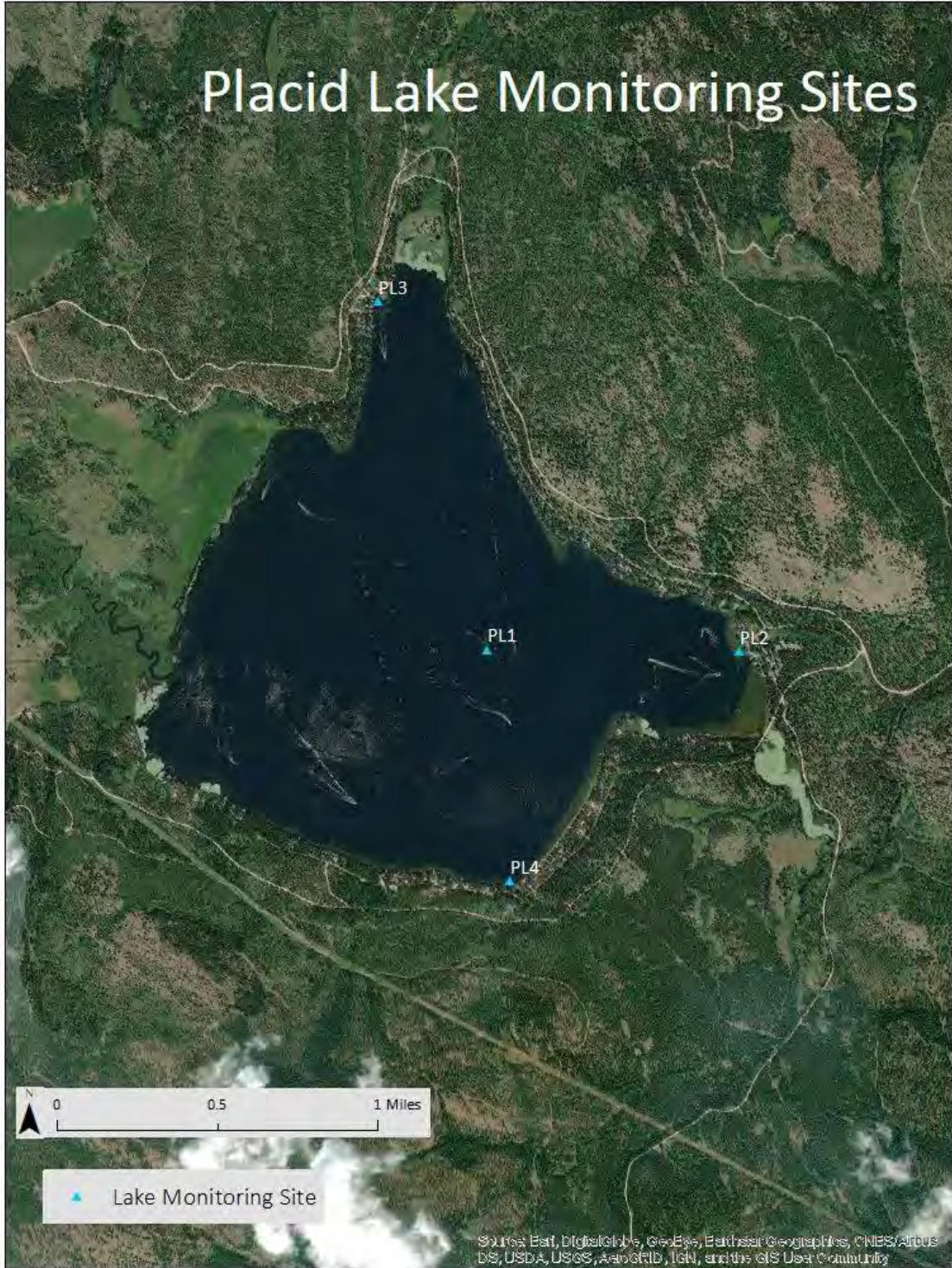
Lake Alva Monitoring Site



Lake Inez Monitoring Sites



Placid Lake Monitoring Sites



Salmon Lake Monitoring Sites



Seeley Lake Monitoring Sites

