

*The Toxicity of the Lotic Water Bodies In the Silverton Caldera  
Concerning the Ability To Support *Hexagenia limbata*, the Giant  
Mayfly*

*By Evan Tweedie*

*Colorado Science and Engineering Fair*

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*Full Report*



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***Abstract***

After I experienced the effects of the Gold King Mine wastewater spill in 2015, I asked the question: which creeks in the Animas River's watershed could lead to its toxicity? I did a previous science fair project where I discovered that the creeks near Silverton were the most toxic.

My science fair project this year is on the toxicity of the lotic water bodies in the Silverton Caldera concerning the ability to support *Hexagenia limbata*, the giant mayfly. In this project, I retrieved water samples from various creeks and rivers before they reached the Animas River. I tested them for their pH, total dissolved solids, turbidity, and dissolved oxygen. I also exposed *Hexagenia limbata*, the giant mayfly to each of these sources and recorded their mortality.

My results showed that Cement Creek and Mineral Creek were the most toxic. They had the highest TDS, lowest pH, and the highest turbidity. These two creeks also had the highest mortality rate in *Hexagenia limbata* populations. Dissolved oxygen did not play a very large factor. Cement Creek and Mineral Creek were the most toxic lotic water bodies and they also had some of the highest concentrations of mine water flowing into them. This helped support my hypothesis. Not only did Cement Creek have the highest toxicity and the concentration of mines in its watershed, it also was the headwaters of the Gold King Mine Spill in 2015.

***Research Paper***

In the morning of August 5, 2015, just north of the town of Silverton, Colorado, The Gold King Mine released up to three million gallons of wastewater rich in iron oxide and zinc into Cement Creek, a tributary of the Animas River. The mix of zinc, iron oxide, and other chemicals in the water turned it an eerie bright orange. The Animas River carried this heavy metal-contaminated water into the San Juan river, which then meets up with the Colorado river in Lake Powell, Utah.

Along this stretch of over 250 miles between Lake Powell and Silverton, there lie many towns and cities that depend on the San Juan and Animas Rivers for clean water. Farmington, Aztec, Durango, and Silverton were affected the most by this disaster. The Four Corners power plant, which is located just outside of Farmington, New Mexico is one facility that depends on the Animas River to function. The plant uses the heat from the burning of coal to boil fresh water into steam, which then spins a turbine to produce electricity. This water comes from Morgan lake, which is fed by the Animas River. If there are any flaws in the water's quality, the plant will not draw any water from Morgan lake and in turn produce no electricity. The city of Farmington depends greatly on the Four Corners power plant for electricity. During the time of this event, which lasted around a week, the plant did not produce any electricity, therefore it was a significant threat to Farmington's power supply.

After its occurrence, the blowout was investigated by the United States Environmental Protection Agency (EPA). The Gold King mine wastewater spill was fixed as well as caused by the EPA. They had previously been damming the water seeping from the GKM. However, when all of this heavily-polluted water became too much for the dam, it burst, flushing all of the water into Cement Creek, where it then met up with the Animas River. This dam failure was partly caused by a long series of problems and minor leaks that had been occurring over the past several decades. However, this was the largest outburst that the GKM had seen yet. Over the years, this water had seeped out little by little, until this particular spill, when it reached its worst.

There are many different measurements that can be used to determine water toxicity. The most well known out of these is pH. pH measures the acidity of a water sample, on a scale of 1-10. A pH of 7.5 is considered safe for most animals. Any pH reading higher than 7.5 is alkaline or base while anything lower than 7.5 is acidic. pH can be measured in three main ways. One simple way to measure pH is using paper coated in pH indicating dye. Another way to measure pH is using a liquid testing kit with pH indicating dye that can be dropped into water to determine its pH based off of the color of the water. Perhaps the most accurate and easiest way of measuring pH is with a digital pH meter.

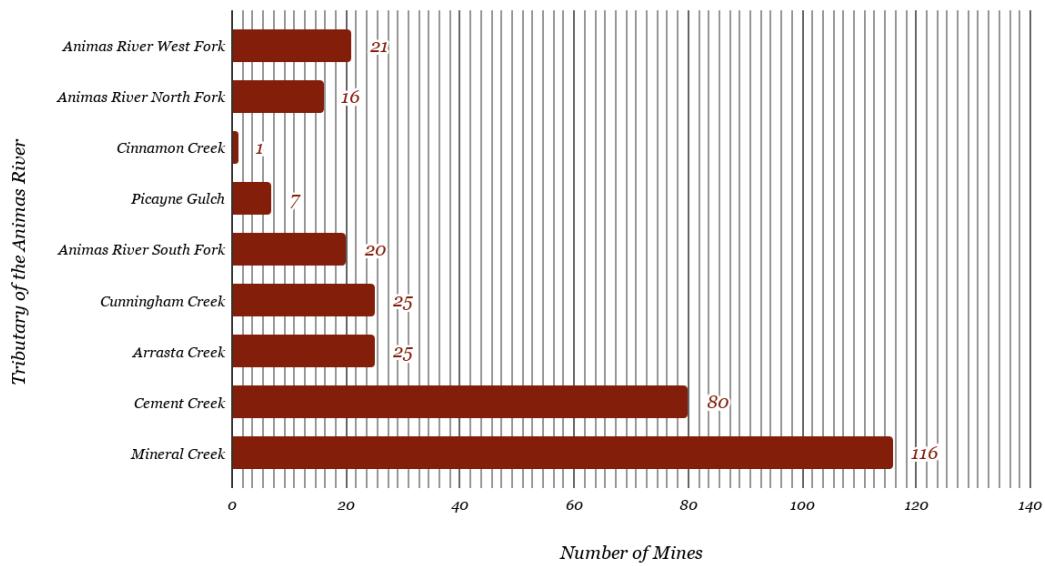
Another measurement that can sometimes be used to determine water is total dissolved solids, or TDS. Total dissolved solids is a measurement of the dissolved combined content of all organic and inorganic substances in a liquid. Though it is considered a primary pollutant, this measurement could or couldn't be used to determine water quality, depending on what the dissolved solids in the water actually are. In the Silverton Caldera, these dissolved solids could be a number of things: salts, heavy metals, minerals, and any other solids that are dissolvable in water. TDS is usually measured in parts per million (ppm) and is usually measured using a TDS meter.

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Turbidity can also sometimes be used in water quality testing. Turbidity is a measurement of the cloudiness or haziness that a liquid has from suspended matter. A high TDS can lead to a high turbidity. The suspended matter that leads to a high turbidity can be the same dissolved content that leads to a high TDS, therefore turbidity in a water source can indicate the same salts, heavy metals, and minerals that TDS helps indicate. Turbidity is usually measured using a special turbidity meter. This meter shoots a beam of light at a liquid sample, and then collects the light on the other side of the sample with a light sensor. If the light sensor detects a lot of light, the sample likely has a low turbidity. Likewise, if the sensor detects very little light, the sample has a high turbidity.

Another way of measuring turbidity is to use a secchi disk, which consists of a large (usually 30 centimeters in diameter) disk with four black and white quadrants. The disk has a weight on the bottom of it and a rope attached to the top of it. It is dropped into a large body of water such as a lake. The user holds on to the string as they lower the secchi disk into the water. Once they cannot see the black and white design on the disk anymore, they mark the spot on the string that was at the water level. Then, they raise the disk out of the water and measure the distance between the disk and the spot that they marked. This measurement is a function of the water's turbidity.

*Number of Mines In the Animas River Watershed*



The table above shows the number of mines in the watersheds of the following creeks and rivers: the Animas River's West, North, and South Forks, Cinnamon Creek, Picayne Gulch, Cunningham Creek, Arrasta Creek, Cement Creek, and Mineral Creek. As you can see, Mineral Creek has the most mines in its watershed out of these nine water sources with a total of 116 mines. Cement Creek also has a large amount of mines in its watershed with a total of 80 mines. On the other hand, Cinnamon Creek with 1 mine and Picayne Gulch with 7 mines have barely any mines in their watershed. The rest of these sample locations have a medium amount of mines in their watershed, ranging from about 16-25 mines.

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***Question***

Which lotic water bodies (creeks and rivers) within the Silverton Caldera may be considered toxic (unable to support animal life)?

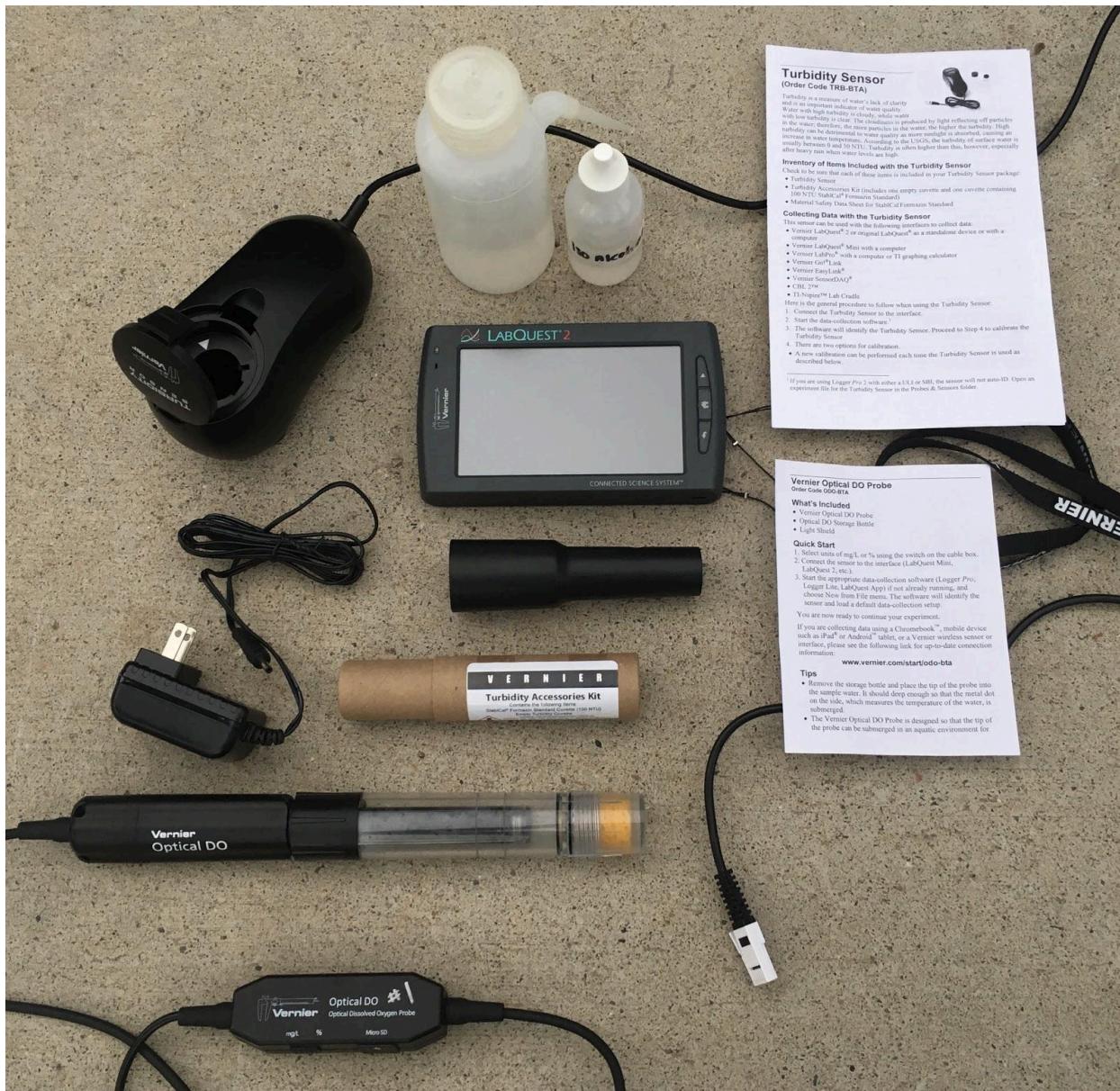
***Hypothesis***

If I test for the pH, TDS, turbidity, and mortality of *Hexagenia limbata* (giant mayfly) in various lotic water samples retrieved throughout the Silverton Caldera, then I will find that the water bodies with more mines in their watershed will prove to be more toxic, because they will contain higher concentrations of heavy metals that could increase the pH, TDS, and turbidity of the water to a point where they may be hazardous to animal health.

## **Materials**

- 400 count - Giant Mayfly Nymphs (*Hexagenia limbata*)
- 10 count - 1 quart glass mason jars
- 30 count - 120-150 millimeter (diameter) sterile petri dishes for use with mayfly larvae
- 10 count - 100 millimeter (diameter) sterile petri dishes for use with microscope
- 3 count - beakers or jars (at least 250 milliliters volume)
- 10 count - 10 milliliter pipettes with cleaning brush or measuring cup with 100 milliliter measurements.
- Measuring tape with measurements in meters (metric) (it should be at least 3 meters long, although the length depends on the depth of the tributaries)
- Pair of tweezers
- Permanent Marker
- Small Labels
- Lint free cloth
- Paper towels
- Small fine mesh net
- Large, shallow rectangular dish
- Isopropyl alcohol
- Distilled (or spring) water
- Sample water - 9 freshwater samples from desired lotic water bodies
- LabQuest (Vernier) with charging cable
- Optical total dissolved oxygen (DO) meter (Vernier) with protective cone
- Turbidity Meter (Vernier)
- Turbidity accessories kit for calibration (Vernier) (includes StablCal formazin standard cuvette and empty turbidity cuvette)
- Digital total dissolved solids and electrical conductivity (TDS & EC) meter (Pancellent)
- pH Meter (Vernier)
- Composition notebook and pen/pencil for collecting data and taking notes
- Secchi disk (if the water is deep enough to where you cannot easily see the bottom)
- Backpack, satchel, or other type of bag for carrying off-hand items
- Chest waders (optional)
- 13 times zoom macro lens (optional)
- Camera (optional, I used the camera on an iPhone 6s)

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(Above) I used various Vernier branded tools and devices in my project such as a LabQuest, Optical Dissolved Oxygen meter and Turbidity sensor. Some of these devices also required materials for calibration.

## **Procedure**

### **Part 1: Gather and Prepare Materials, Calibrate the Devices at Home**

1. Gather the materials listed above.
2. Calibrate the turbidity sensor if needed. Recalibration is required during long periods of inactivity or very frequent use, as well as if the testing accuracy requirement is very high. Calibrate the turbidity sensor using the turbidity accessories kit (includes StablCal formazin standard cuvette and empty turbidity cuvette). See **Appendix II** for instructions on calibration.
3. Label the length of the rope on the secchi disk using the measuring tape and a permanent marker. Measure 0.5 meters, 1 meter, 1.5 meters, and so on from the actual black and white secchi disk (measure the length by the centimeter for more accurate results). The extent of the measurements varies greatly on the depth and murkiness of the source water, and the secchi disk may not even be usable if the water is clear and shallow. Please note that secchi disks are usually not used in lotic (flowing) water bodies, but I will attempt to use one for the sake of this project.

### **Part 2: Record Data in the Field**

1. Go to the various tributaries of the Animas River in the Silverton Caldera and complete the following while wading into each tributary:
  - a. Wade out into the deepest part of the creek or river (also known as the thalweg of the river) if it is safe. Then, walk upstream for at least a meter, so that you are not collecting any water that was mixed with sediment from the creekbed.
  - b. Fill one of the 1-quart jars from upstream of you, and then dump the water downstream of you. This helps to clean out the jars before filling them. Do this about 3 times.
  - c. Fill one of the mason jars with the water that is coming from upstream of you. Hold the jar level in the water and place the cap onto it, making sure that the water is not shaken up to prevent any air bubbles from forming in the jar. Then, carefully place it in your backpack, satchel, or other type of bag. Please note that it is best to only go into the creek or river once, so that as little sediment as possible is stirred up from the bottom of the creek.
  - d. Take out the empty cuvette from the Vernier turbidity accessories kit. Then, fill it and then dump it downstream of you about 3 times, just as you had with the 1-quart jar. Fill the jar at the thalweg and carefully place it in your bag, without shaking it up.
  - e. Now, measure the pH of the water. Take out the pH probe and plug it into the Vernier LabQuest device. Then, turn on the LabQuest, place the probe into the thalweg (it is waterproof up to the device itself), and take the reading. The LabQuest has a few nifty tools in it that can allow you to store the data in a graph or table. Save the data on the LabQuest, record data in the creek, or read off to a partner at the shore. Remove the probe from the water and rinse it downstream with distilled water. Then, carefully dry off the probe with a paper towel and disconnect it from the LabQuest.
  - f. Next, measure the total dissolved solids (TDS) in the water as well as the temperature in Celsius. As with the pH meter, remove the cap, place the TDS & EC meter into the water at the thalweg (if possible), and turn on the probe. Then record data in the creek or read it off to a partner at the shore. Again, make sure that the probe has stopped fluctuating before recording data. Turn it off, remove it from the stream, rinse it off with distilled water, dry it using a paper towel, place the cap onto it, and place it back into your pack as you had done with the pH meter.

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- g. Then, measure the dissolved oxygen in the water. To do this, take out the Vernier dissolved oxygen meter, plug it into the Vernier LabQuest device, and place the protective cone onto it. Then, turn on the LabQuest, place the probe into the thalweg, and take the reading. Then, save the data on the LabQuest, record data in the creek, or read off to a partner at the shore. Remove the probe from the water, remove the protective cone and rinse it downstream like you did with the pH and TDS & EC meters with distilled water. Then, carefully dry off the probe with a paper towel and disconnect it from the LabQuest.
- h. If the creek or river is deep enough, measure the water's turbidity with a secchi disk. Take out the secchi disk and lower the disk into the water with the string until the black and white pattern is not visible. At this point, measure the length of the string using the measurements from before. Make sure you are taking the measurement with the blue foam handle at the water's surface. Record data while in the creek or tell a partner at the shore. Place everything back into your bag and walk back to shore.

2. When you get back to shore, complete the following steps:
  - a. Measure for the turbidity. Take out the cuvette that you filled earlier as well as the Vernier turbidity probe and the LabQuest. Plug the turbidity probe into the LabQuest and open the cylindrical compartment on the probe. Make sure the turbidity probe is set on a flat, level surface. Open the cuvette, dump out a small amount of the water until it reaches the small white line on the side of the jar, and place the cap back on the cuvette. Take out the lint-free cloth and Isopropyl alcohol and put a few drops of the alcohol onto the cloth. Then, wipe off the cuvette with the cloth and place it into the cylindrical compartment in the turbidity probe, holding it by the cap. Make sure that the white arrows on the cuvette and in the probe line up. Then, turn on the LabQuest and record data. Finally, put everything away in your bag.
  - b. Number and label the mason jar with the coordinates, date, and time in which they were collected as well as the tributary that they were collected from. Then, cap the jar, place it in your bag, and go to the next tributary.
3. Complete each of these steps about 9 times, once for each source in one day if possible, to assure that the time is somewhat constant.
4. Fill one of the jars with distilled water or spring water and label it 'Control'. Complete this step only once.

**Part 3: Experiment with *Hexagenia limbata* Nymphs (Giant Mayfly Larvae)**

1. Obtain the mayfly nymphs. They can be purchased online on online bait shops (I ordered mine from *The Reel Thing Bait and Tackle*) or in local bait shops and fisheries. They are often called 'wrigglers' because of their swimming techniques (especially in the Wisconsin area). I ordered 400 of the larvae, assuming that 300 would survive the overnight shipping, but this project could definitely be tested on smaller or larger scales.
2. Keep them in a long, shallow rectangular tank if necessary before experimentation. The reason that the tank should be long and shallow rather than tall and deep is because the larvae will want to sit at the surface, and they will not be able to do so in a tall tank. The mayflies do not need much water at this stage. In fact, they should not have much water at all, to prevent them from dying of exhaustion (With too much water, they will swim themselves to death). The tank does not need to be oxygenated, but will need cleaned at some point if they are being kept at this stage for a long period of time.
3. Complete the following steps for the mayfly nymphs during experimentation:
  - a. Label the 10 pipettes and 30 petri dishes with the corresponding number to the source in which they will be used for.

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- b. Using the pipettes, fill each petri dish with 100 milliliters of the source water that was collected in the 1-quart jars. There will be 3 petri dishes for each source and 10 sources total. Shake up the jars directly before pouring the water into the petri dishes, to dissolve any sediment that may have settled on the bottom of the jar.
- c. Using the small fine mesh net, transfer 10 mayfly nymphs into each petri dish. There will be 30 mayfly nymphs being tested per source. Do not use the tweezers to handle them when they are alive, for this may stress them out or severely injure them.
- d. Record how many are still alive after the first 30 minutes, 1 hour, 3 hours, 12 hours, and 24 hours. After that, continue to check them every 24 hours or so for a maximum of 7 days until none remain alive. (I introduced the nymphs to the water at 5:00 PM and checked them at 5:30, 6:30, 9:30, and so on). Record data for every check, including the number of mayflies that had died and any observations that are worth noting.
- e. As soon as any mayfly nymphs die, use the tweezers to remove them. Then, place the deceased mayflies in a 100 millimeter petri dish that corresponds with the sample that they are from. After using the tweezers to remove individuals from one sample, rinse the tweezers with distilled water before using them in another source's water to prevent contamination. Mayfly nymphs can be disposed of in the trash after they die.

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(Above) My mayfly experimentation setup took place in my garage, which had an average temperature of 60-70 degrees fahrenheit (15.5-21.1 degrees celsius) while this testing took place. I had three trials for each sample and about 10 mayflies per trial for a total of 30 trials and 300 mayflies.

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## ***Risk Assessment***

### ***Hazards***

- Various Chemicals in the creek water
- Risk of mold growth in samples
- Potential to handle chemicals for calibrations
- Wading in the water to collect samples

### ***Risks***

- Very low or high pH in water may pose health risk
- Risk of mold growth in petri dishes with mayflies
- Chemicals can be dangerous to handle and cause health issues
- Getting swept away in a river during collection of the water samples or falling in and getting cold and possibly leading to hypothermia.

### ***Precautions***

- Minimal handling of the collected water samples and use of pipette to avoid contact.
- Samples will be checked regularly and dead mayflies will be removed immediately upon discovery and will be left to dry to prevent mold growth.
- Minimal handling of calibration chemicals using gloves.
- Only wading into the river if it is safe and not very fast. Using chest waders to avoid getting wet during cold winter months. Collecting water samples near roadways near a car; if I fall in and get cold there is a quick way to warm up.

### ***Disposal Procedures***

- Disposing of creek water down the sink or toilet and not on the land outside.
- Petri dishes showing any signs of mold growth will be disposed of immediately without opening.
- All insects will be dead prior to disposal to avoid spreading a potentially invasive species.

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## **Results / Analysis**

### **Part 1: Field Measurements**

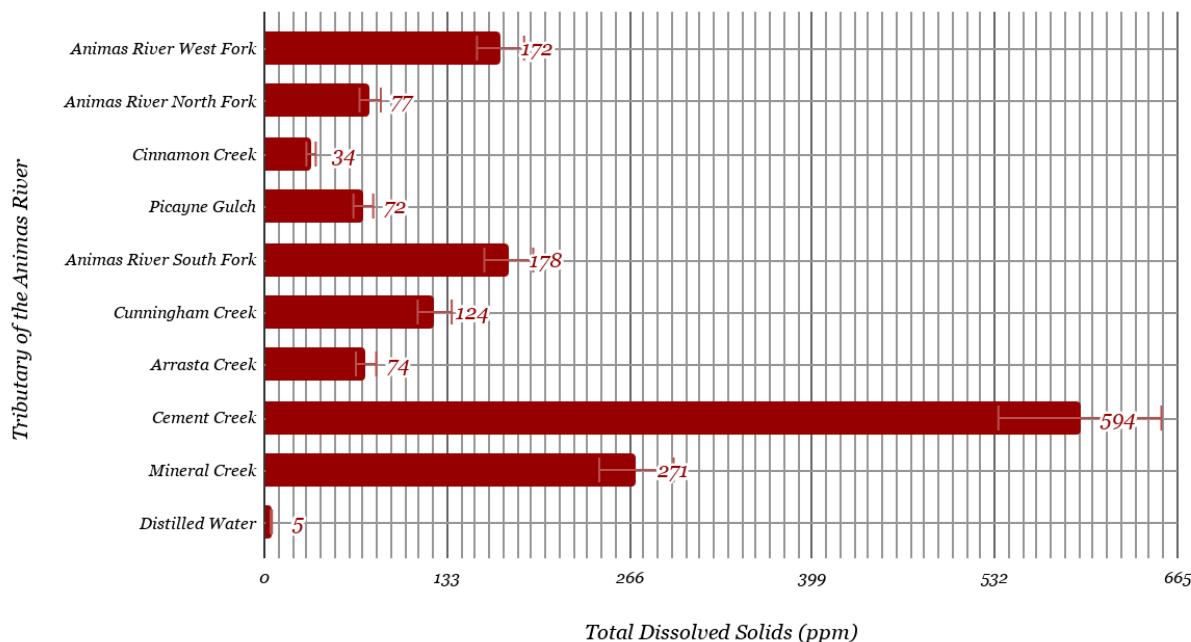
**Table 1: General Measurements**

| Sample Number | Sample Location         | TDS (ppm) | Turbidity (NTU) | Dissolved Oxygen (mg/L) | pH  | Latitude (°North) | Longitude (°West) | Temp. (°Celcius) | Elevation (in meters and feet)   |
|---------------|-------------------------|-----------|-----------------|-------------------------|-----|-------------------|-------------------|------------------|----------------------------------|
| 1             | Animas River West Fork  | 172 ppm   | 25.5 NTU        | 8.66 mg/L               | 5.8 | 37.93196 °N       | -107.57085 °W     | 0.0 °C           | 11,161 feet<br>3,401.8728 meters |
| 2             | Animas River North Fork | 77 ppm    | 2.7 NTU         | 7.95 mg/L               | 6.3 | 37.93267 °N       | -107.56956 °W     | 0.0 °C           | 11,208 feet<br>3,416.1984 meters |
| 3             | Cinnamon Creek          | 34 ppm    | 8.5 NTU         | 9.73 mg/L               | 7.0 | 37.92597 °N       | -107.56278 °W     | -4.6 °C          | 11,018 feet<br>3,358.2864 meters |
| 4             | Picayne Gulch           | 72 ppm    | 10.0 NTU        | 8.34 mg/L               | 7.9 | 37.91140 °N       | -107.55596 °W     | 0.0 °C           | 10,661 feet<br>3,249.4728 meters |
| 5             | Animas River South Fork | 178 ppm   | 10.3 NTU        | 9.55 mg/L               | 7.5 | 37.87960 °N       | -107.56766 °W     | 6.8 °C           | 9,896 feet<br>3,016.3008 meters  |
| 6             | Cunningham Creek        | 124 ppm   | 6.9 NTU         | 7.44 mg/L               | 7.6 | 37.83504 °N       | -107.59519 °W     | 8.6 °C           | 9,705 feet<br>2,958.0840 meters  |
| 7             | Arrasta Creek           | 74 ppm    | 6.7 NTU         | 7.54 mg/L               | 7.6 | 37.82641 °N       | -107.62552 °W     | 7.9 °C           | 9,575 feet<br>2,918.4600 meters  |
| 8             | Cement Creek            | 594 ppm   | 23.4 NTU        | 8.05 mg/L               | 3.8 | 37.81456 °N       | -107.66132 °W     | 6.6 °C           | 9,345 feet<br>2,848.3560 meters  |
| 9             | Mineral Creek           | 271 ppm   | 33.8 NTU        | 8.67 mg/L               | 5.0 | 37.80269 °N       | -107.67250 °W     | 5.4 °C           | 9,262 feet<br>2,823.0576 meters  |
| 10            | Distilled Water         | 5 ppm     | 0 NTU           | 7.3 mg/L                | 7.5 | N/A               | N/A               | 25 °C            | N/A                              |

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**Bar Chart 1: Total Dissolved Solids Levels in Various Sample Locations (ppm)**

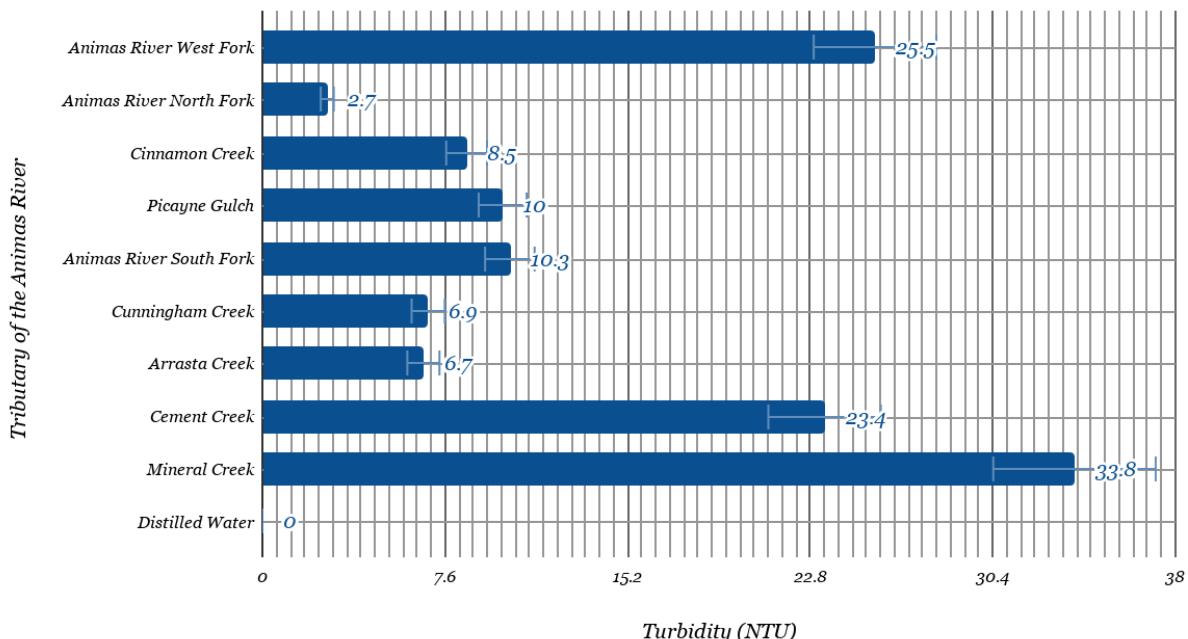
**Total Dissolved Solid Levels In Various Sample Locations (ppm)**



The bar chart above shows the levels of total dissolved solids (TDS) in the water from each of the sample sites that I visited in parts per million (ppm). These measurements were taken in the creeks themselves rather than in jars or at home to prevent them from being inaccurate due to the settling of particles. These dissolved solids could consist of a number of things, including salts, heavy metals, and other minerals. As you can see, Cement Creek has a TDS of 594 parts per million, the highest of all of the samples. It is followed by Mineral Creek with a TDS of 271 parts per million. Cinnamon Creek has the lowest TDS out of all of the creek samples with a TDS of 34 parts per million. It is followed by Picayne Gulch, Arrasta Creek, and the Animas River's North Fork, with a TDS range of 72 to 77 parts per million. Cunningham Creek and the Animas River's West and South Forks had a moderate TDS with a range of 124 to 178 parts per million. If one were to look at a map showing all of the mines near these creeks, they would find that Cinnamon Creek, which flows west into the Animas River near a town called Animas Forks, has barely any mines in its headwaters. It also has a lot of dense vegetation around its banks. The same can be found at the headwaters of Cunningham and Arrasta Creek, which are located on the southern end of the Silverton Caldera, just due east of the town of Silverton. Cement Creek, on the other hand, has many mines in its headwaters, including the famed Gold King Mine, which was the route of a massive mine spill in 2015. Mineral Creek is significantly larger than the rest of the other tributaries and flows into the Animas River near Silverton from the West. It has a significant amount of mines in its headwaters as well. The Animas River's West and North Forks are located in the town of Animas Forks and have some mines in their headwaters, but not many. The Animas River's South Fork in Eureka, however, has quite a bit of mining activity. This shows that the creeks with more mining activity in their headwaters tend to have more total dissolved solids in their water.

**Bar Chart 2 : Turbidity Levels in Various Samples (NTU)**

**Turbidity Levels In Various Sample Locations (NTU)**

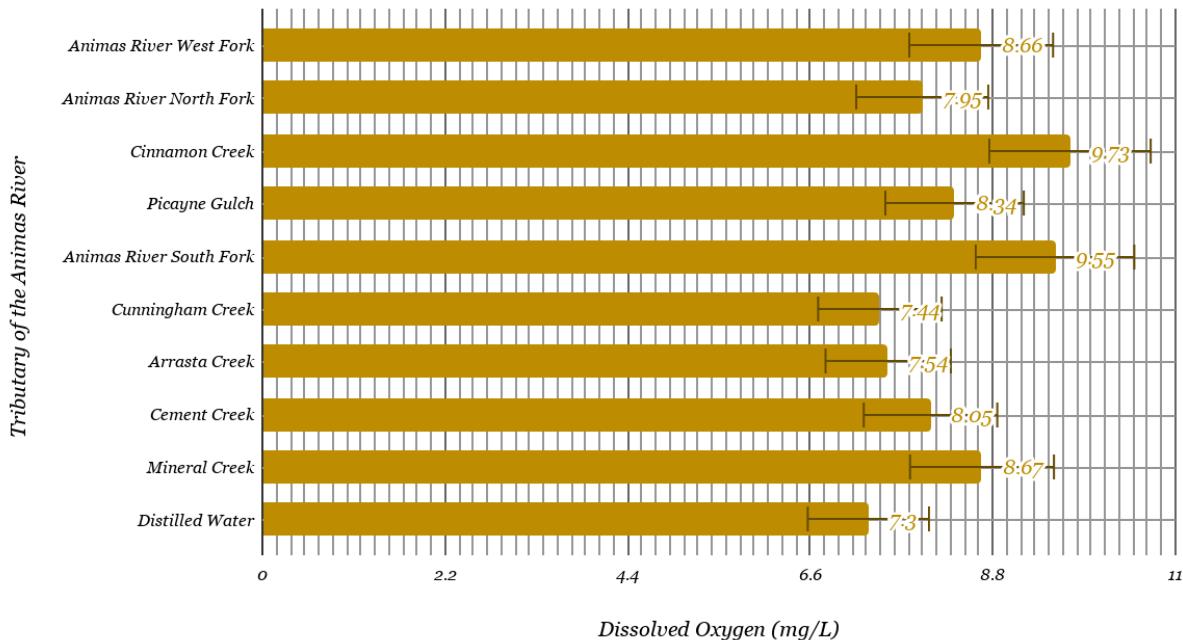


The bar chart above shows the turbidity levels of the water from each of the sample sites that I visited in nephelometric turbidity units (NTU). These measurements were taken in the creeks themselves rather than in jars or at home to prevent them from being inaccurate due to the settling of particles. Turbidity can be thought of as the murkiness of the water or how much light can pass through it. As you can see, Mineral Creek has the highest turbidity of 33.8 nephelometric turbidity units. It is followed by the Animas River's West Fork, with a turbidity of 25.5 nephelometric turbidity units. Cement Creek has the third highest turbidity number with a turbidity of 23.4 nephelometric turbidity units. The creek sample with the lowest turbidity is The Animas River's North Fork, with a turbidity of 2.7 nephelometric turbidity units. The rest of the samples have a relatively low turbidity, with a range of 6.7 to 10.3 nephelometric turbidity units. This graph somewhat correlates with the previous graph on TDS levels, but instead of Cement Creek having the highest turbidity, Mineral Creek does, which has the second highest TDS level. Another difference is that the Animas River's West Fork has a surprisingly high turbidity level compared to its TDS level, a moderate 172 parts per million. I also found it interesting how much the turbidity differed between the Animas River's West and North Forks, because both had a very similar level of TDS and both were very close to each other. I expected that they would not differ very much because of their close proximity to each other.

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**Bar Chart 3: Dissolved Oxygen in Various Sample Locations (mg/L)**

*Dissolved Oxygen Levels In Various Sample Locations (mg/L)*



The bar chart above shows the levels of dissolved oxygen in the water from each of the sample sites that I visited in milligrams per liter (mg/L). These measurements were taken in the creeks themselves rather than in jars or at home to prevent them from being inaccurate. These measurements were taken mostly on behalf of the *H. limbata* larvae, in case I saw a correlation between lack of dissolved oxygen and mayfly mortality. As it turns out, there was a significant correlation between mayfly larvae deaths and lack of dissolved oxygen. After 24 hours, the distilled water sample had the fourth to most deaths. It also had the lowest dissolved oxygen level out of all of the samples. Cinnamon Creek had the second to least amount of deaths and the highest dissolved oxygen level.

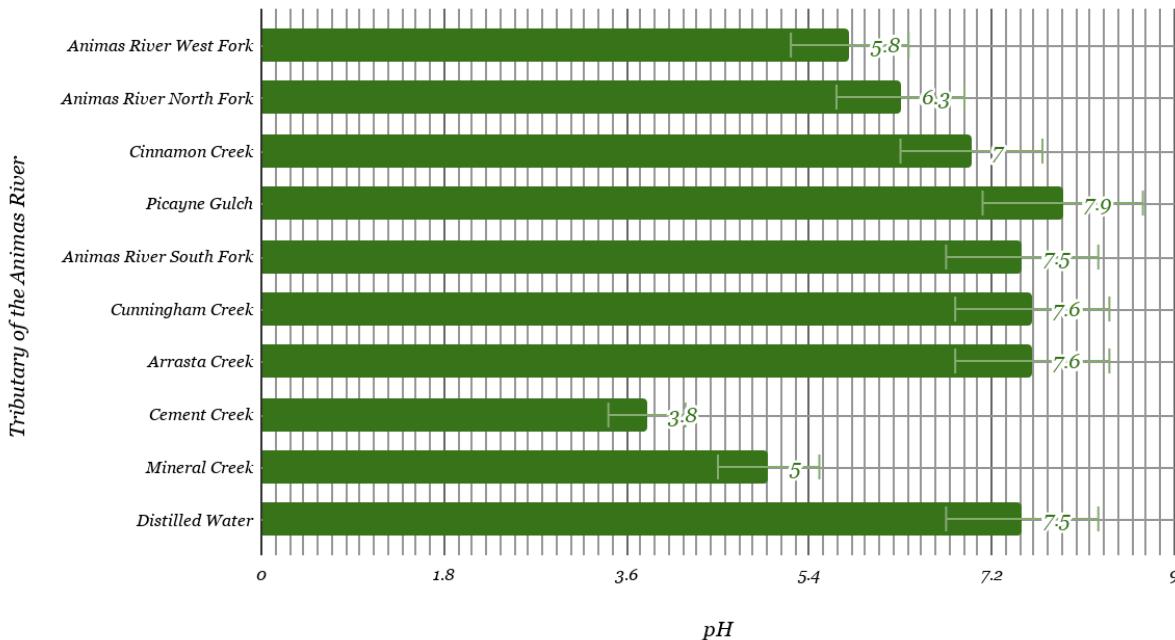
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**Bar Chart 4: pH Levels in Various Sample Locations**

*pH Levels In Various Sample Locations (pH)*

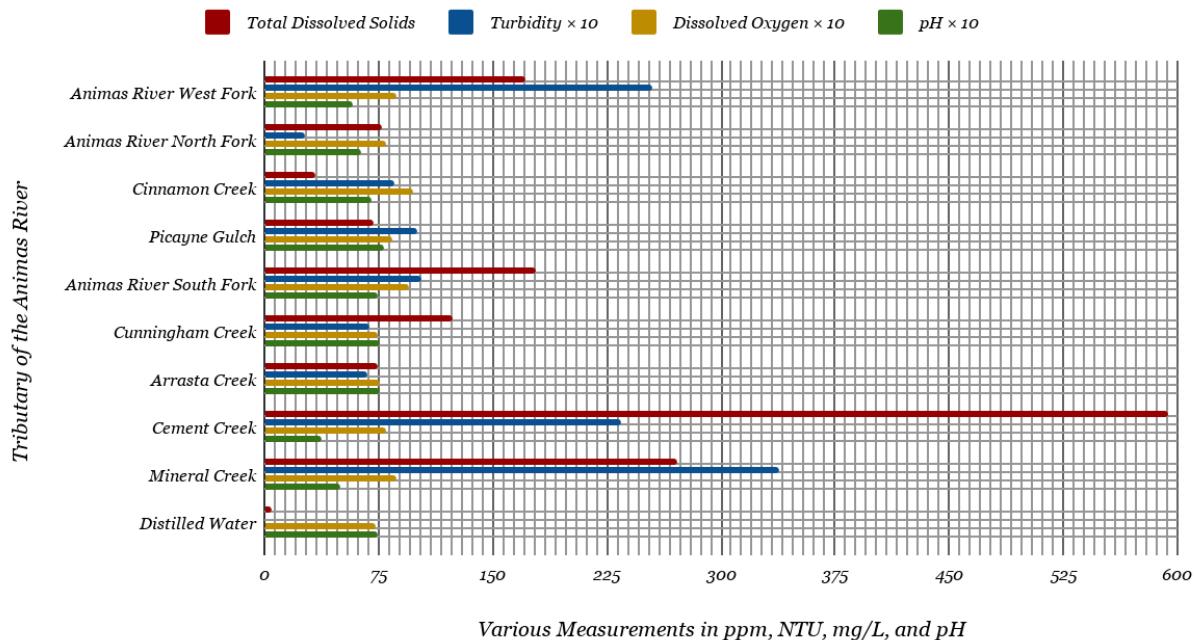


The bar chart above shows the pH of the water at each of the sample locations that I visited. These measurements were meant to be taken in the creeks themselves because temperature can slightly alter a pH reading. However, because the pH meter that I had planned on using had worn out, I had to take the measurements at home, where I tested the samples with a separate pH meter. As you can see, most of the sample's pH readings did not vary much, except for Cement Creek and Mineral Creek, with very acidic pH levels of 3.8 and 5. The West and North Forks of the Animas River were acidic as well, with levels of 5.8 and 6.3. The other samples were at safe levels, varying from 7 to 7.9 in pH. Distilled water has a pH of 7.5, which is usually considered the safest pH level for water consumption.

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**Bar Chart 5: All Field Measurements (Not to Scale)**

**All Field Measurements (Not To Scale)**



The bar chart above compares the total dissolved solids, turbidity, dissolved oxygen, and pH of each of the sample locations that I visited. This graph is a combination of the previous graphs and is for comparative purposes only. It is not to scale because the turbidity, dissolved oxygen, and pH levels have been increased by a factor of 10 so that any differences between the samples will be more visible. The results for total dissolved solids levels did not change. These measurements also vary in format. Total dissolved solids is measured in ppm, turbidity in NTU, dissolved oxygen in mg/L, and pH in standard pH format.

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**Part 2: Field Observations and Notes**

I collected all of my samples on November 10, 2019 in the Silverton Caldera. The sources that I checked include the Animas River (West, North, and South Forks), Cinnamon Creek, Picayne Gulch, Cunningham Creek, Arrasta Creek, Cement Creek, and Mineral Creek. The weather for that day was mostly sunny, with a few altostratus clouds in the sky.

**Table 2: Field Observations and Notes**

| Sample Number | Sample Location         | Time of Arrival | Observations (physical)  | Observations (geological)  | Observations (biological)   | Observations (human impact)   |
|---------------|-------------------------|-----------------|--|--|---|---|
| 1             | Animas River West Fork  | 10:25 AM        | Mostly frozen, fast flowing river, blanketed by snow. Medium light levels.   | Igneous intrusive rounded stones on creek bed, with somewhat whitish coating when exposed to air. (calcium?) Water with light blue tinge, deepest parts reaching a meter at most.  | Medium-high diversity, close to tundra, lots of krumholtz (Engelmann spruce and Subalpine fir) and a few Bebb willows along banks. All annuals and biennials are long gone and covered by snow. | Medium impact, old mill ruins with white, gray, and orange tailing piles trailing very close to the river. Dirt roads as well as bridge crossing. |
| 2             | Animas River North Fork | 11:00 AM        | Mostly frozen, fast flowing river, blanketed by snow. Significantly less snowpack than WF. Medium-high light levels.                 | Igneous intrusive rounded stones on banks and creek bed, with whitish coating when exposed to air. (calcium?) Water with light blue tinge, deepest parts reaching half a meter at most. More rocky outcrops than WF.   | Medium-high diversity, close to tundra, lots of krumholtz (Engelmann spruce and Subalpine fir) and many Bebb willows along banks. Some pussytoes on rocky bits (perennial).                     | Low impact, single dirt road crossing river.  |
| 3             | Cinnamon Creek          | 11:30 AM        | Mostly frozen, medium-slow flowing creek, blanketed by snow. Lower light levels. Ice needed to be broken to access the river easily. | Primarily igneous intrusive rounded stones on creek bed, no obvious precipitation of minerals on rocks or dissolved in the water. Very shallow, reaching around one fourth of a meter where measurements were taken. Cliff on the southern edge of creek, igneous intrusive. | High diversity, subalpine with full growth trees including Bebb willow, as well as Engelmann spruce and Subalpine fir in rocky areas.   | Low impact, single dirt road crossing creek.  |
| 4             | Picayne Gulch           | 12:00 AM        | Medium-slow flowing creek with little ice and snow. Direct sunlight  | Igneous intrusive gravel like pebbles covering the creek bed along with larger igneous intrusive boulders. The   | Medium-high diversity, mostly small, deciduous vegetation on the shore such as Bebb willow  | Low impact, single gravel road crossing creek.  |

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|   |                         |          |  |  |  |  |
|---|-------------------------|----------|--|--|--|--|
|   |                         |          | with high light levels.  | pebbles coating the creek bed had a greenish-yellow tinge, possibly from algal growth or precipitation of minerals. Very shallow, reaching around one fourth of a meter where measurements were taken.   | with some blue spruce krumholtz away from the creek. The pebbles coating the creek bed had a greenish-yellow tinge, possibly from algal growth. Lots of green plate lichen and moss was present.   |  |
| 5 | Animas River South Fork | 12:40 AM | Mostly frozen, fast flowing river, blanketed in a thin layer of snow. Medium-high light levels.      | Igneous intrusive rounded stones in creek bed, no obvious precipitation of minerals on rocks or dissolved in the water. Most of the creek seemed shallow, around one fifth of a meter where measurements were taken. The river was coming out of a deep basalt gorge and had some mining activity. | Medium-high diversity, mostly small, deciduous vegetation on the shore such as Bebb willow with some blue spruce krumholtz away from the creek. Some organic material was present in the river itself, including algae on some of the rocks. | Medium-high impact, old mine shaft due south of the river and a mill due north of it that could be leaching mineral and heavy metal deposits. Samples of rhodonite, hematite, quartz, pumice, chalcopyrite, and iron pyrite were found in the mill ruins. Located in the ghost town of Eureka. |
| 6 | Cunningham Creek        | 1:20 PM  | Medium-fast flowing creek with no ice or snow. Direct sunlight with high light levels.               | Igneous intrusive rounded stones in creek bed with possible precipitation of calcium and an orangish-green mineral when out of water. Most of the creek was shallow, around one fourth of a meter where measurements were taken.   | Medium-high diversity, mostly small, deciduous vegetation on the shore such as Bebb willow with some blue spruce krumholtz away from the creek.  | Medium-low impact, small bridge crossing the creek and a dirt road parallel to it.   |
| 7 | Arrasta Creek           | 1:40 PM  | Medium-fast flowing creek with very little ice and no snow. Indirect sunlight with low light levels. | Igneous intrusive rounded stones in creek bed with a greenish tinge. Most of the creek was shallow in a deep ditch, around one fourth of a meter where measurements were taken.  | High diversity with subalpine fir, Engelmann spruce, and smaller deciduous vegetation on the creek banks. Some blue spruce was present as well.  | Low impact, small dirt road parallel to the creek.   |
| 8 | Cement Creek            | 1:55 PM  | Fast flowing creek with no ice or snow. Direct sunlight with high light levels.                      | Igneous intrusive rounded stones on creek bed, very obvious precipitation of what looks to be zinc and iron oxide deposits on rocks and dissolved in the water. The creek was very shallow, around one fifth of a meter where measurements were  | Low diversity with only a few deciduous shrubs on the creek banks.   | Very high impact with roads and buildings surrounding the creek and a bridge crossing over it. A significant amount of litter was present as well. Located in the town of Silverton.   |

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|   |               |         |   | taken.   |  |   |
|---|---------------|---------|---|--|--|---|
| 9 | Mineral Creek | 2:20 PM | Fast flowing creek with very little ice and no snow present. Direct sunlight with low light levels. | Igneous intrusive rounded stones on creek bed, very obvious precipitation of what looks to be zinc and iron oxide deposits on rocks and dissolved in the water. The creek was very shallow, around one fifth of a meter where measurements were taken. | Medium-low diversity with a few blue spruce, Engelmann spruce, and subalpine fir on the creek banks. Some deciduous vegetation was also present. | Medium-high impact with a dirt road leading up to the creek and an old, rusty building on one side of it. |

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**Part 3: Giant Mayfly (*Hexagenia limbata*) Investigation**

**Table 3: *Hexagenia limbata* Mortality Over Time In Animas River West Fork Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1            | 0        | 0      | 0      | 0       | 0       | 1       | 1       | 1       | 0       | 1       | 1       | 1       | 6               | 4              |
| 2            | 0        | 0      | 0      | 0       | 0       | 3       | 1       | 0       | 0       | 1       | 2       | 0       | 7               | 3              |
| 3            | 0        | 0      | 0      | 0       | 2       | 4       | 0       | 0       | 0       | 0       | 1       | 1       | 8               | 2              |

**Table 4: *Hexagenia limbata* Mortality Over Time In Animas River North Fork Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1            | 0        | 0      | 0      | 0       | 1       | 5       | 2       | 0       | 0       | 0       | 0       | 0       | 8               | 2              |
| 2            | 0        | 0      | 0      | 0       | 1       | 3       | 1       | 1       | 0       | 0       | 1       | 0       | 7               | 3              |
| 3            | 0        | 0      | 0      | 0       | 2       | 2       | 1       | 0       | 0       | 0       | 0       | 0       | 5               | 5              |

**Table 5: *Hexagenia limbata* Mortality Over Time In Cinnamon Creek Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1            | 0        | 0      | 0      | 0       | 2       | 1       | 0       | 0       | 1       | 1       | 1       | 0       | 6               | 4              |
| 2            | 0        | 0      | 0      | 0       | 2       | 0       | 2       | 0       | 0       | 1       | 0       | 0       | 5               | 5              |
| 3            | 0        | 0      | 0      | 0       | 2       | 1       | 0       | 1       | 1       | 0       | 1       | 0       | 6               | 4              |

**Table 6: *Hexagenia limbata* Mortality Over Time In Picayne Gulch Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1            | 0        | 0      | 0      | 0       | 2       | 1       | 2       | 0       | 1       | 0       | 1       | 0       | 7               | 3              |
| 2            | 0        | 0      | 0      | 0       | 1       | 2       | 0       | 2       | 1       | 0       | 2       | 0       | 8               | 2              |
| 3            | 0        | 0      | 0      | 0       | 2       | 3       | 0       | 3       | 0       | 0       | 0       | 0       | 8               | 2              |

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**Table 7: *Hexagenia limbata* Mortality Over Time In Animas River South Fork Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1            | 0        | 0      | 0      | 0       | 2       | 1       | 1       | 0       | 0       | 0       | 0       | 4               | 6              |
| 2            | 0        | 0      | 0      | 0       | 2       | 2       | 1       | 0       | 0       | 0       | 0       | 3               | 7              |
| 3            | 0        | 0      | 0      | 0       | 0       | 2       | 3       | 2       | 0       | 0       | 0       | 7               | 3              |

**Table 8: *Hexagenia limbata* Mortality Over Time In Cunningham Creek Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |   |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|---|
| 1            | 0        | 0      | 0      | 0       | 1       | 4       | 1       | 0       | 0       | 1       | 0       | 0               | 7              | 3 |
| 2            | 0        | 0      | 0      | 0       | 2       | 4       | 1       | 0       | 0       | 0       | 1       | 0               | 8              | 2 |
| 3            | 0        | 0      | 0      | 0       | 2       | 3       | 1       | 1       | 0       | 0       | 0       | 1               | 8              | 2 |

**Table 9: *Hexagenia limbata* Mortality Over Time In Arrasta Creek Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |   |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|---|
| 1            | 0        | 0      | 0      | 0       | 0       | 3       | 0       | 0       | 0       | 0       | 2       | 0               | 5              | 5 |
| 2            | 0        | 0      | 0      | 0       | 1       | 1       | 1       | 0       | 0       | 0       | 0       | 0               | 3              | 7 |
| 3            | 0        | 0      | 0      | 0       | 5       | 2       | 1       | 0       | 0       | 0       | 0       | 0               | 8              | 2 |

**Table 10: *Hexagenia limbata* Mortality Over Time In Cement Creek Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |   |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|---|
| 1            | 0        | 0      | 0      | 0       | 2       | 3       | 1       | 1       | 0       | 1       | 2       | 0               | 10             | 0 |
| 2            | 0        | 0      | 0      | 0       | 1       | 2       | 1       | 2       | 3       | 1       | 0       | 0               | 10             | 0 |
| 3            | 0        | 0      | 0      | 0       | 3       | 1       | 2       | 1       | 0       | 0       | 0       | 0               | 7              | 3 |

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**Table 11: Hexagenia limbata Mortality Over Time In Mineral Creek Sample Water**

| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1            | 0        | 0      | 0      | 0       | 1       | 2       | 1       | 3       | 0       | 1       | 0       | 1               | 9              |
| 2            | 0        | 0      | 0      | 0       | 0       | 1       | 3       | 0       | 1       | 0       | 1       | 0               | 6              |
| 3            | 0        | 0      | 0      | 1       | 2       | 4       | 0       | 1       | 0       | 1       | 0       | 0               | 9              |

**Table 12: Hexagenia limbata Mortality Over Time In Distilled Water**

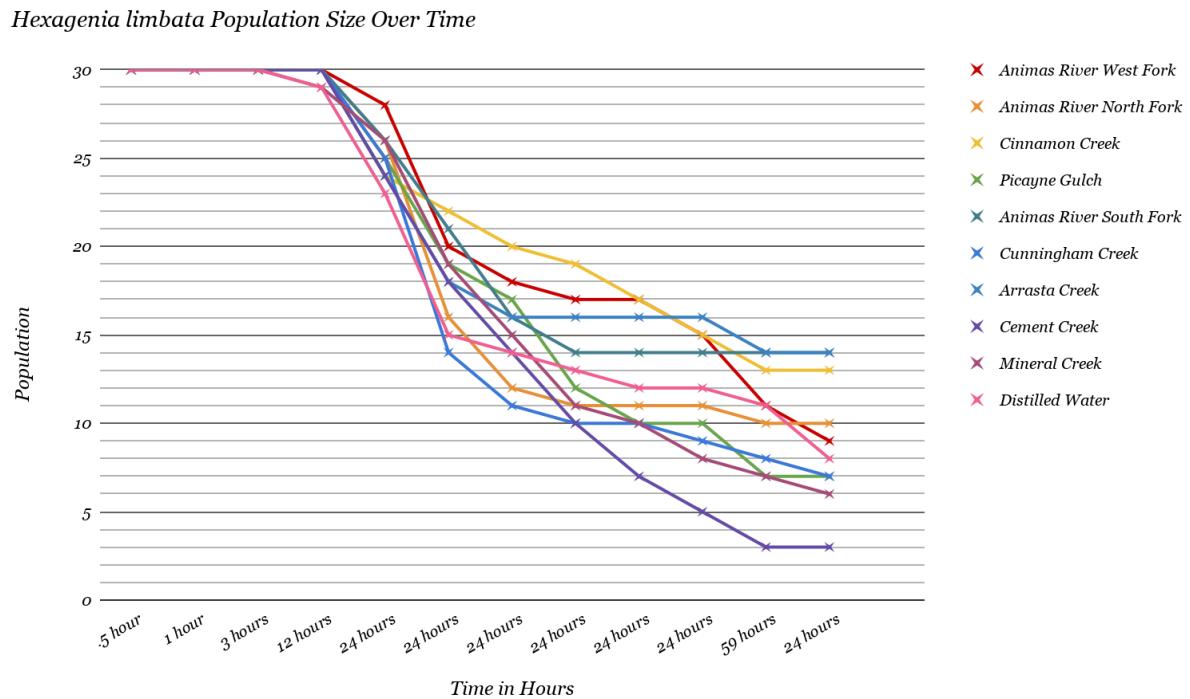
| Trial Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|--------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1            | 0        | 0      | 0      | 0       | 3       | 2       | 0       | 0       | 0       | 0       | 0       | 5               | 5              |
| 2            | 0        | 0      | 0      | 1       | 1       | 1       | 1       | 1       | 0       | 0       | 2       | 8               | 2              |
| 3            | 0        | 0      | 0      | 0       | 2       | 5       | 0       | 0       | 0       | 0       | 1       | 1               | 9              |

**Table 13: Total Mortality From Hexagenia limbata Investigation In Various Samples**

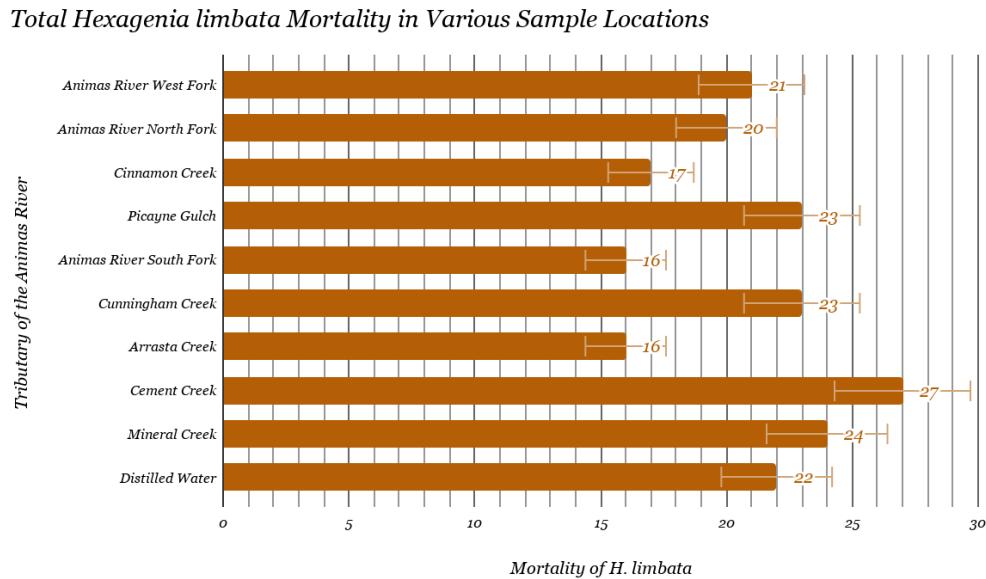
| Sample Number | 0.5 hour | 1 hour | 3 hour | 12 hour | 24 hour | 59 hour | 24 hour | Total Mortality | Total Survival |
|---------------|----------|--------|--------|---------|---------|---------|---------|---------|---------|---------|---------|-----------------|----------------|
| 1             | 0        | 0      | 0      | 0       | 2       | 8       | 2       | 1       | 0       | 2       | 4       | 2               | 21             |
| 2             | 0        | 0      | 0      | 0       | 4       | 10      | 4       | 1       | 0       | 0       | 1       | 0               | 20             |
| 3             | 0        | 0      | 0      | 0       | 6       | 2       | 2       | 1       | 2       | 2       | 2       | 0               | 17             |
| 4             | 0        | 0      | 0      | 0       | 5       | 6       | 2       | 5       | 2       | 0       | 3       | 0               | 23             |
| 5             | 0        | 0      | 0      | 0       | 4       | 5       | 5       | 2       | 0       | 0       | 0       | 0               | 16             |
| 6             | 0        | 0      | 0      | 0       | 5       | 11      | 3       | 1       | 0       | 1       | 1       | 1               | 23             |
| 7             | 0        | 0      | 0      | 0       | 6       | 6       | 2       | 0       | 0       | 0       | 2       | 0               | 16             |
| 8             | 0        | 0      | 0      | 0       | 6       | 6       | 4       | 4       | 3       | 2       | 2       | 0               | 27             |
| 9             | 0        | 0      | 0      | 1       | 3       | 7       | 4       | 4       | 1       | 2       | 1       | 1               | 24             |
| 10            | 0        | 0      | 0      | 1       | 6       | 8       | 1       | 1       | 1       | 0       | 1       | 3               | 22             |
| Total         | 0        | 0      | 0      | 2       | 47      | 69      | 29      | 20      | 9       | 9       | 17      | 7               | 207            |
|               |          |        |        |         |         |         |         |         |         |         |         |                 | 93             |

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**Line Graph 1: Giant Mayfly Population Size Over Time In Various Samples**



**Bar Chart 6: Total Mayfly Mortality In Various Sample Locations**



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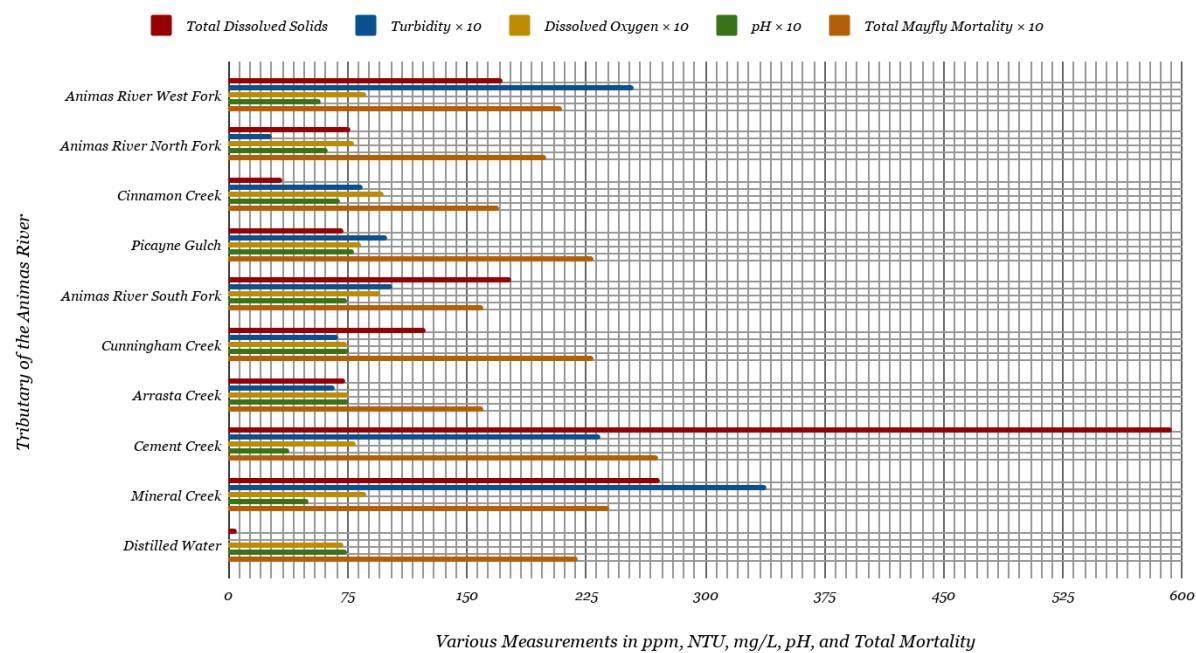
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The line graph and bar chart above show *Hexagenia limbata* mortality/population changes in each of the sample locations that I visited. As you can see, in the first 16.5 hours, there was nearly no mortality in any of the samples. Then, the mayfly populations all began to drop at a steady rate. About 48 hours later, most of the populations began to level out. However, the populations in some of the samples such as Mineral Creek and Cement Creek continued to drop. This helped differentiate each of the samples from the rest. After the full 243.5 hours was completed, Cement Creek had the highest mortality of 27 deaths. Mineral Creek had the second highest mortality with a total of 24 deaths. Picayne Gulch and Cunningham Creek also had a high mortality rate with a total of 23 deaths, but not significantly different from Distilled Water.

**Bar Chart 7: All Field Measurements With Total *Hexagenia limbata* Mortality (Not To Scale)**

*All Field Measurements With Total *Hexagenia limbata* Mortality (Not To Scale)*



The bar chart above is a copy of **Bar Chart 5** with total mayfly mortality added in. It is for comparison only and the total mayfly mortality value has been increased by a factor of 10.

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## **Conclusion**

In conclusion, the most toxic water bodies with the highest TDS, highest turbidity, lowest pH, and highest mayfly mortality include those of which have the most mines in their watersheds. The two most toxic samples that I tested were Cement Creek with 80 mines and Mineral Creek with 116 mines. This information shows that my hypothesis was proven.

According to my data on TDS content in each of these creeks, the sample with the most total dissolved solids was Cement creek with 594 parts per million. The sample with the second most total dissolved solids was Mineral creek with 271 parts per million. As for turbidity, Mineral creek had the highest turbidity level with 33.8 NTU. The creek with the second highest turbidity level was The Animas River's West Fork, with a turbidity of 25.5 NTU. The creek with the lowest pH was Cement creek, with a pH of 3.8. It was followed by Mineral creek, with a pH of 5. In the study with *H. limbata*, Cement Creek had the highest total mortality with a total of 27 deaths. It was followed by Mineral creek with a total of 24 deaths.

In the future, if this project were to be continued, a greater sample size could be used for *H. limbata* nymphs or I could use *Daphnia magna* (water fleas) again as I did with last year's project. They work as a good biological indicator for water toxicity just like *H. limbata*, making them a good choice for this type of project. I could also test for other factors such as a river's flow rate and volume, which is important for this type of experiment. This type of research is important because so many people depend on clean water from the Animas River, which is greatly influenced by the creeks and rivers in its upper watershed.

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*The Toxicity of the Lotic Water Bodies In the Silverton Caldera Concerning the Ability To Support*

*Hexagenia limbata, the Giant Mayfly*

*By Evan Tweedie*

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I would also like to thank my environmental science teacher, Sasha Creeden for peer reviewing my project and helping provide me with some of my materials.

## ***Appendix***

### ***Appendix I: The Mines of Each Source's Watershed***

#### ***Animas River West Fork***

| Mine Name                       | Primary Commodities      | Secondary Commodities    |
|---------------------------------|--------------------------|--------------------------|
| Columbus Gold Mine              | Gold                     | Copper, Silver, and Zinc |
| San Antonio Mine                | Gold                     | Copper, Lead, and Silver |
| Frisco Tunnel                   | Copper, Gold, and Lead   | NA                       |
| Bagley Gold Mine                | Gold                     | Silver                   |
| Little Ida Mine                 | Lead                     | Copper, Gold, and Silver |
| Burrows Lead Mine               | Lead and Zinc            | NA                       |
| Vermillion Tunnel               | Gold                     | Copper, Lead, and Silver |
| Vermillion Zinc Mine            | Gold, Lead, and Silver   | NA                       |
| Silver Cord Mine                | Lead                     | Zinc                     |
| Indian Chief Mine               | Gold                     | Lead, Silver, and Zinc   |
| Mountain Queen Mine             | Lead                     | Gold, Silver, and Zinc   |
| Custer Lead-Silver Mine         | Lead and Silver          | NA                       |
| Black Diamond Copper Occurrence | Copper, Lead, and Silver | NA                       |
| Evening Star Mine               | Copper, Lead, and Zinc   | Gold and Silver          |
| Independence Gold Mine          | Gold                     | Copper, Lead, and Silver |
| Silver Queen Mine               | Copper, Lead, and Zinc   | NA                       |
| Sound Democrat Mine             | Gold, Lead, and Silver   | NA                       |
| Hidden Treasure Mine            | Gold and Silver          | NA                       |

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|                              |                        |    |
|------------------------------|------------------------|----|
| Silver Queen Lead Occurrence | Lead and Silver        | NA |
| Silver Queen Lead Occurrence | Copper, Lead, and Zinc | NA |
| Treasure Mountain Mine       | Gold and Silver        | NA |

***Animas River North Fork***

| Mine Name                   | Primary Commodities      | Secondary Commodities |
|-----------------------------|--------------------------|-----------------------|
| Unnamed Zinc Mine           | Zinc                     | Gold and Lead         |
| Upper Uncompahgre Mines     | Tungsten                 | NA                    |
| Blacksmith Tungsten Mine    | Tungsten                 | NA                    |
| Lucky Jack Lead Mine        | Copper, Gold, and Lead   | NA                    |
| Unnamed Zinc Mine           | Zinc                     | Gold and Lead         |
| Ben Butler Gold-Silver Mine | Copper, Gold, and Lead   | NA                    |
| Unnamed Zinc Mine           | Zinc                     | Gold and Lead         |
| Sewell Lead-Silver Mine     | Lead and Silver          | NA                    |
| Red Cloud Silver Mine       | Copper, Gold, and Lead   | NA                    |
| Uncompahgre Chief Mine      | Silver and Tungsten      | Lead and Zinc         |
| Early Bird Gold Mine        | Gold and Silver          | NA                    |
| Dewitt Lead-Silver Mine     | Copper, Lead, and Silver | NA                    |
| Unnamed Zinc Mine           | Zinc                     | Gold                  |
| Unnamed Zinc Mine           | Zinc                     | Gold and Lead         |
| Hermes Group                | Copper, Gold, and Lead   | NA                    |
| Unnamed Zinc Group          | Zinc                     | Gold and Lead         |

***Cinnamon Creek***

| Mine Name              | Primary Commodities | Secondary Commodities |
|------------------------|---------------------|-----------------------|
| Detroit Hollister Mine | Gold                | Silver                |

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***Picayne Gulch***

| Mine Name              | Primary Commodities    | Secondary Commodities |
|------------------------|------------------------|-----------------------|
| Toltec Lead Mine       | Lead                   | Copper and Zinc       |
| Unnamed Zinc Mine      | Zinc                   | Lead                  |
| Scotia Lead Occurrence | Copper, Lead, and Zinc | NA                    |
| Sandiago Gold Mine     | Gold                   | Silver                |
| Blanchard Placer       | Gold                   | NA                    |
| Golden Fleece Mine     | Gold                   | NA                    |
| San Juan Queen Mine    | Lead, Silver, and Zinc | NA                    |

***Animas River South Fork***

| Mine Name                    | Primary Commodities      | Secondary Commodities  |
|------------------------------|--------------------------|------------------------|
| Ransome Lode                 | Copper, Silver, and Zinc | NA                     |
| Plain Streak Mine            | Silver                   | Copper, Gold, and Lead |
| Unnamed Zinc Mine            | Zinc                     | Copper, Gold, and Lead |
| Montezuma Gold-Silver Mine   | Gold and Silver          | Copper, Lead, and Zinc |
| Unnamed Zinc Occurrence      | Zinc                     | Gold and Lead          |
| Silver Ledge Tungsten Mine   | Tungsten                 | NA                     |
| Natalie Tungsten Mine        | Tungsten                 | NA                     |
| Kittimac Zinc Mine           | Copper, Lead, and Zinc   | NA                     |
| Unnamed Zinc Mine            | Zinc                     | Copper, Gold, and Lead |
| Unnamed Zinc Mine            | Zinc                     | Gold and Lead          |
| Aztec Gold Mine              | Gold                     | Lead, Silver, and Zinc |
| Silver Queen Mine            | Lead and Zinc            | NA                     |
| Silver Queen Lead Occurrence | Copper, Lead, and Zinc   | NA                     |

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|                            |                        |                          |
|----------------------------|------------------------|--------------------------|
| Terry Tunnel               | Copper                 | Lead and Zinc            |
| American Tunnel            | Gold                   | Copper, Lead, and Silver |
| Ben Franklin Mine          | Lead, Silver, and Zinc | NA                       |
| Grivitzia Lead-Silver Mine | Copper, Gold, and Lead | NA                       |
| George Washington Mine     | Gemstone               | NA                       |
| Unnamed Gold Mine          | Gold                   | Lead, Silver, and Zinc   |
| Belle Creole Mine          | Gold                   | Lead, Silver, and Zinc   |

***Cunningham Creek***

| Mine Name                       | Primary Commodities    | Secondary Commodities    |
|---------------------------------|------------------------|--------------------------|
| Old Hundred Mine                | Lead                   | Gold, Silver, and Zinc   |
| Niegold Group                   | Gold                   | Copper, Lead, and Silver |
| Gary Owens Mine                 | Lead                   | Gold, Silver, and Zinc   |
| Unnamed Zinc Mine               | Zinc                   | Copper, Gold, and Lead   |
| Buffalo Boy Mine                | Gold and Silver        | Copper, Lead, and Zinc   |
| Antiperiodic Lead-Silver Mine   | Gold, Lead, and Silver | NA                       |
| Galena Mountain Group           | Lead                   | Gold, Silver, and Zinc   |
| Osceola Pride Copper Occurrence | Copper, Gold, and Lead | NA                       |
| Pride of the West Lead Mine     | Gold, Lead, and Silver | NA                       |
| Philadelphia Lead Mine          | Lead and Silver        | NA                       |
| Little Fanny Mine               | Silver                 | Lead                     |
| Green Mountain Mine             | Lead and Silver        | NA                       |
| Pride of the West Mine          | Copper, Gold, and Lead | NA                       |
| Green Mountain Mine             | Copper, Lead, and Zinc | NA                       |
| Unnamed Zinc Group              | Zinc                   | Gold and Lead            |

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|                        |                 |                          |
|------------------------|-----------------|--------------------------|
| North Star Silver Mine | Lead and Silver | NA                       |
| Dives Lead-Silver Mine | Lead and Silver | NA                       |
| Shanandoah Dives Mill  | Sulfur-Pyrite   | NA                       |
| Trilby Group           | Gold            | Copper, Lead, and Silver |
| Unnamed Zinc Group     | Zinc            | Copper, Gold, and Lead   |
| Lookout Gold Mine      | Gold            | Copper, Lead, and Silver |
| Highland Mary Mine     | Uranium         | NA                       |
| Unnamed Zinc Mine      | Zinc            | Copper, Gold, and Lead   |
| Unnamed Zinc Mine      | Zinc            | Copper, Gold, and Lead   |
| Gold Lake Mine         | Silver          | Copper, Gold, and Lead   |

***Arrasta Creek***

| Mine Name             | Primary Commodities      | Secondary Commodities  |
|-----------------------|--------------------------|------------------------|
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Little Nation Mine    | Copper, Gold, and Lead   | NA                     |
| Unnamed Zinc Mine     | Zinc                     | Copper and Gold        |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Big Giant Copper Mine | Copper, Lead, and Silver | NA                     |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Mayflower Gold Mine   | Gold                     | Lead and Silver        |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Little Giant Mine     | Gold                     | NA                     |

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|                                  |                        |                          |
|----------------------------------|------------------------|--------------------------|
| Unity Tunnel                     | Gold                   | Copper, Lead, and Silver |
| Unnamed Zinc Mine                | Zinc                   | Copper, Gold, and Lead   |
| Shenandoah Lead Mine             | Gold, Lead, and Zinc   | NA                       |
| Shenandoah-Dives Zinc Occurrence | Copper, Gold, and Lead | NA                       |
| Silver Lake Group                | Copper, Lead, and Zinc | NA                       |
| Silver Lake Mine                 | Lead and Silver        | NA                       |
| Iowa Lead Mine                   | Lead and Silver        | NA                       |
| Royal Tiger Mine                 | Lead and Silver        | NA                       |
| Unnamed Zinc Group               | Zinc                   | Copper, Gold, and Lead   |
| Unnamed Zinc Mine                | Zinc                   | Copper, Gold, and Lead   |
| Unnamed Zinc Mine                | Zinc                   | Copper, Gold, and Lead   |
| Unnamed Zinc Mine                | Zinc                   | Copper, Gold, and Lead   |
| Buckeye Mine                     | Lead and Silver        | Manganese                |

***Cement Creek***

| Mine Name             | Primary Commodities      | Secondary Commodities  |
|-----------------------|--------------------------|------------------------|
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Unnamed Silver Mine   | Silver                   | Lead and Zinc          |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Irene Group           | Lead                     | Copper and Silver      |
| Boston Lead-Zinc Mine | Copper, Gold, and Lead   | NA                     |
| Porcupine Lode        | Copper, Lead, and Silver | NA                     |
| Unnamed Zinc Mine     | Zinc                     | Copper, Gold, and Lead |
| Golden Hub Mine       | Tungsten                 | NA                     |
| Delano Tungsten Mine  | Tungsten                 | NA                     |

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|                             |                        |                                     |
|-----------------------------|------------------------|-------------------------------------|
| Omaha Zinc Mine             | Zinc                   | Copper, Gold, and Lead              |
| Omaha Zinc Mine             | Zinc                   | Copper, Gold, and Lead              |
| Omaha Tungsten Mine         | Tungsten               | Copper, Lead, and Silver            |
| Anglo-Saxon Mine            | Tungsten               | Fluorine-Fluorite, Gold, and Silver |
| Ariadne Tungsten Mine       | Tungsten               | NA                                  |
| Sunshine Tungsten Mine      | Tungsten               | NA                                  |
| Unnamed Tungsten Mine       | Tungsten               | NA                                  |
| Cement Bog Iron Mine        | Iron                   | NA                                  |
| Minnesota Gulch Mine        | Zinc                   | Copper, Gold, and Lead              |
| Minnesota Zinc Occurrence   | Zinc                   | Copper, Gold, and Lead              |
| Unnamed Zinc Mine           | Zinc                   | Copper, Gold, and Lead              |
| Tin Cup Tungsten Occurrence | Tungsten               | NA                                  |
| Belcher Lead Mine           | Lead and Zinc          | Copper and Silver                   |
| Conyer Tungsten Mine        | Tungsten               | NA                                  |
| Graham Mine                 | Lead, Silver, and Zinc | Copper and Gold                     |
| Crevice Lead Mine           | Lead                   | Silver and Zinc                     |
| Unnamed Zinc Mine           | Zinc                   | Copper, Gold, and Lead              |
| Unnamed Zinc Mine           | Zinc                   | Copper, Gold, and Lead              |
| Unnamed Zinc Mine           | Zinc                   | Copper, Gold, and Lead              |
| Hoosier Boy Mine            | Zinc                   | Copper, Gold, and Lead              |
| Unnamed Zinc Group          | Zinc                   | Copper, Gold, and Lead              |
| Kansas City Mine            | Silver                 | Copper, Lead, and Zinc              |
| Graham Silver Mine          | Silver                 | NA                                  |
| Galena Queen Mine           | Copper, Lead, and Zinc | Gold and Silver                     |

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|                             |                        |                          |
|-----------------------------|------------------------|--------------------------|
| Henrietta Mine              | Copper, Lead, and Zinc | Gold and Silver          |
| Elk Silver Mine             | Silver                 | Iron and Lead            |
| Lark Zinc Mine              | Copper, Lead, and Zinc | NA                       |
| Joe and John Lower Mine     | Copper, Gold, and Lead | NA                       |
| Unnamed Zinc Mine           | Zinc                   | Copper, Gold, and Lead   |
| Unnamed Zinc Mine           | Zinc                   | Copper, Gold, and Lead   |
| Silver Cloud Mine           | Copper, Lead, and Zinc | NA                       |
| Dawn of Day Mine            | Tungsten               | NA                       |
| Burns Group                 | Lead                   | Copper, Gold, and Silver |
| Unnamed Zinc Mine           | Gold, Lead, and Silver | NA                       |
| Unnamed Zinc Mine           | Zinc                   | Copper, Gold, and Lead   |
| Evaline Tungsten Occurrence | Tungsten               | NA                       |
| Minnesota Tungsten Mine     | Tungsten               | NA                       |
| First Chance Mine           | Tungsten               | NA                       |
| Reed Tunnel                 | Gold                   | Lead, Silver, and Zinc   |
| Unnamed Gold Mine           | Gold, Lead, and Silver | NA                       |
| Little Mack Mine            | Gold                   | Copper, Lead, and Silver |
| Gold King Mill Placer       | Gold and Silver        | NA                       |
| Webster Silver Mine         | Silver                 | NA                       |
| Dawn of Day Mine            | Tungsten               | NA                       |
| Silver Ledge Mine           | Tungsten               | Gold, Lead, and Silver   |
| Gypsy Claim                 | Tungsten               | NA                       |
| Big Colorado Mine           | Tungsten               | NA                       |
| Unnamed Zinc Group          | Zinc                   | Copper, Gold, and Lead   |

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|                                |                          |                             |
|--------------------------------|--------------------------|-----------------------------|
| Gold Thread Tungsten Mine      | Tungsten                 | NA                          |
| Gladstone Gold Mine            | Gold                     | Lead, Silver, and Zinc      |
| Silver Bay Mine                | Copper, Gold, and Lead   | NA                          |
| Blackhawk Gold-Lead Mine       | Gold, Lead, and Zinc     | Copper and Silver           |
| Minnehaha Mine                 | Copper, Lead, and Silver | NA                          |
| Minnehaha Lead Mine            | Lead and Zinc            | NA                          |
| Lead Carbonate Mine            | Lead and Zinc            | Copper, Silver, and Uranium |
| Blackhawk Uranium Mine         | Uranium                  | NA                          |
| Bismark Tungsten Mine          | Tungsten                 | NA                          |
| Lead Carbonate Mine            | Lead                     | Zinc                        |
| Benitoite Lead Mine            | Gold, Lead, and Silver   | NA                          |
| Mockingbird Lead Mine          | Lead                     | Gold, Silver, and Zinc      |
| Red and Bonita Mine            | Tungsten                 | NA                          |
| Eagle Mountain Lead Occurrence | Copper, Lead, and Zinc   | NA                          |
| Adams Lode                     | Tungsten                 | NA                          |
| Pittsburg Lead Mine            | Lead                     | Zinc                        |
| Gold King Mine                 | Gold, Lead, and Silver   | Copper, Tungsten, and Zinc  |
| Mogul Gold Mine                | Gold                     | Copper, Lead, and Silver    |
| Adelphin Gold Mine             | Gold                     | NA                          |
| Red Rogers Mine                | Lead                     | Silver and Zinc             |
| Rose Tungsten Mine             | Tungsten                 | NA                          |
| Queen Anne Gold-Silver Mine    | Gold, Lead, and Silver   | NA                          |
| Columbia Silver Mine           | Lead and Silver          | NA                          |

***Mineral Creek***

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| Mine Name                           | Primary Commodities        | Secondary Commodities    |
|-------------------------------------|----------------------------|--------------------------|
| Mineral Creek Gold Mine             | Gold                       | Copper, Lead, and Silver |
| Gladstone Silver Mine               | Silver                     | Copper, Gold, and Lead   |
| Sultan Mountain Tungsten Occurrence | Tungsten                   | NA                       |
| Victoria Gold Mine                  | Gold                       | Lead, Silver, and Zinc   |
| North Star Lead Mine                | Lead, Silver, and Tungsten | Copper, Gold, and Zinc   |
| Belcer Silver Mine                  | Copper, Lead, and Silver   | NA                       |
| Anvil Mountain South Mine           | Gold                       | NA                       |
| Unnamed Zinc Mine                   | Zinc                       | Copper, Gold, and Lead   |
| Jones Mill                          | Gold                       | Silver                   |
| Unnamed Molybdenum Occurrence       | Molybdenum                 | NA                       |
| Putnam Basin Mine                   | Gold                       | NA                       |
| Silverheels Silver Mine             | Silver                     | Copper, Gold, and Lead   |
| Zuni Lead Mine                      | Lead                       | Copper and Silver        |
| Saratoga Gold Mine                  | Gold                       | Lead, Silver, and Zinc   |
| Black Diamond Mine                  | Molybdenum                 | NA                       |
| Unnamed Lead Mine                   | Lead                       | Copper, Gold, and Silver |
| Unnamed Lead Mine                   | Lead                       | Zinc                     |
| Waco Silver Mine                    | Silver                     | Zinc                     |
| Ensle Tunnel                        | Gold                       | NA                       |
| Burbank Gold Mine                   | Gold                       | NA                       |
| Unnamed Gold Mine                   | Gold                       | NA                       |
| Unnamed Gold Occurrence             | Gold and Silver            | NA                       |
| Unnamed Gold Occurrence             | Gold and Silver            | NA                       |

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|                                |                 |                          |
|--------------------------------|-----------------|--------------------------|
| Golden Gate Mine               | Silver          | Lead and Zinc            |
| Mineral Creek Bog Iron Deposit | Iron            | NA                       |
| Webster Gold Mine              | Gold            | Copper, Lead, and Silver |
| Lucy Gold Mine                 | Gold            | Copper, Lead, and Silver |
| Diamond Gold Mine              | Gold            | Silver                   |
| Columbus-Grandview Gold Mine   | Gold            | Copper, Lead, and Silver |
| Last Hope and Grandview Group  | Silver          | Gold                     |
| Kinney Tunnel                  | Zinc            | Lead and Silver          |
| Unnamed Gold Occurrence        | Gold            | Silver                   |
| Bandora Group                  | Gold            | Copper, Lead, and Silver |
| Unnamed Gold Occurrence        | Gold            | Silver                   |
| Unnamed Gold Occurrence        | Gold            | Silver                   |
| Unnamed Lead Mine              | Lead            | Copper and Zinc          |
| Unnamed Gold Mine              | Gold            | Copper and Silver        |
| Unnamed Gold Mine              | Gold            | NA                       |
| Unnamed Silver Occurrence      | Gold and Silver | NA                       |
| Big Three Mine                 | Gold            | Copper, Lead, and Silver |
| Rolling Mountain Mine          | Gold            | Copper, Lead, and Silver |
| Unnamed Gold Mine              | Gold            | NA                       |
| Unnamed Lead Mine              | Lead            | Silver                   |
| Moly Gulch Mine                | Lead            | Molybdenum               |
| Liberty Bond Creek Mine        | Molybdenum      | NA                       |
| Brobdignag Silver Mine         | Silver          | Lead                     |
| Unnamed Gold Mine              | Gold            | NA                       |

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|                           |                        |                          |
|---------------------------|------------------------|--------------------------|
| Unnamed Gold Mine         | Gold                   | NA                       |
| Unnamed Gold Mine         | Gold                   | NA                       |
| Brooklyn Gold Mine        | Copper, Gold, and Lead | NA                       |
| Unnamed Gold Mine         | Gold                   | Silver and Zinc          |
| Unnamed Zinc Mine         | Zinc                   | Silver                   |
| Mountain View Claims      | Lead                   | Copper, Gold, and Silver |
| Unnamed Lead Mine         | Lead                   | Copper, Silver, and Zinc |
| Unnamed Lead Mine         | Lead                   | Silver                   |
| Zinc-King Mine            | Lead                   | Zinc                     |
| Bonner Gold Mine          | Gold                   | Copper, Lead, and Silver |
| Unnamed Zinc Mine         | Zinc                   | NA                       |
| Unnamed Gold Mine         | Gold                   | NA                       |
| Unnamed Silver Mine       | Silver                 | Lead                     |
| Unnamed Silver Mine       | Silver                 | Lead and Zinc            |
| Unnamed Lead Mine         | Lead                   | Silver and Zinc          |
| Unnamed Gold Mine         | Gold                   | NA                       |
| Unnamed Gold Mine         | Gold                   | NA                       |
| Freida Silver Mine        | Silver                 | NA                       |
| Unnamed Lead Mine         | Lead                   | Silver                   |
| Unnamed Zinc Mine         | Zinc                   | Copper, Gold, and Lead   |
| Unnamed Lead Mine         | Lead                   | Silver and Zinc          |
| Unnamed Gold Mine         | Gold                   | NA                       |
| Unnamed Gold Mine         | Gold                   | NA                       |
| Alto Juan Gold Occurrence | Gold and Silver        | NA                       |

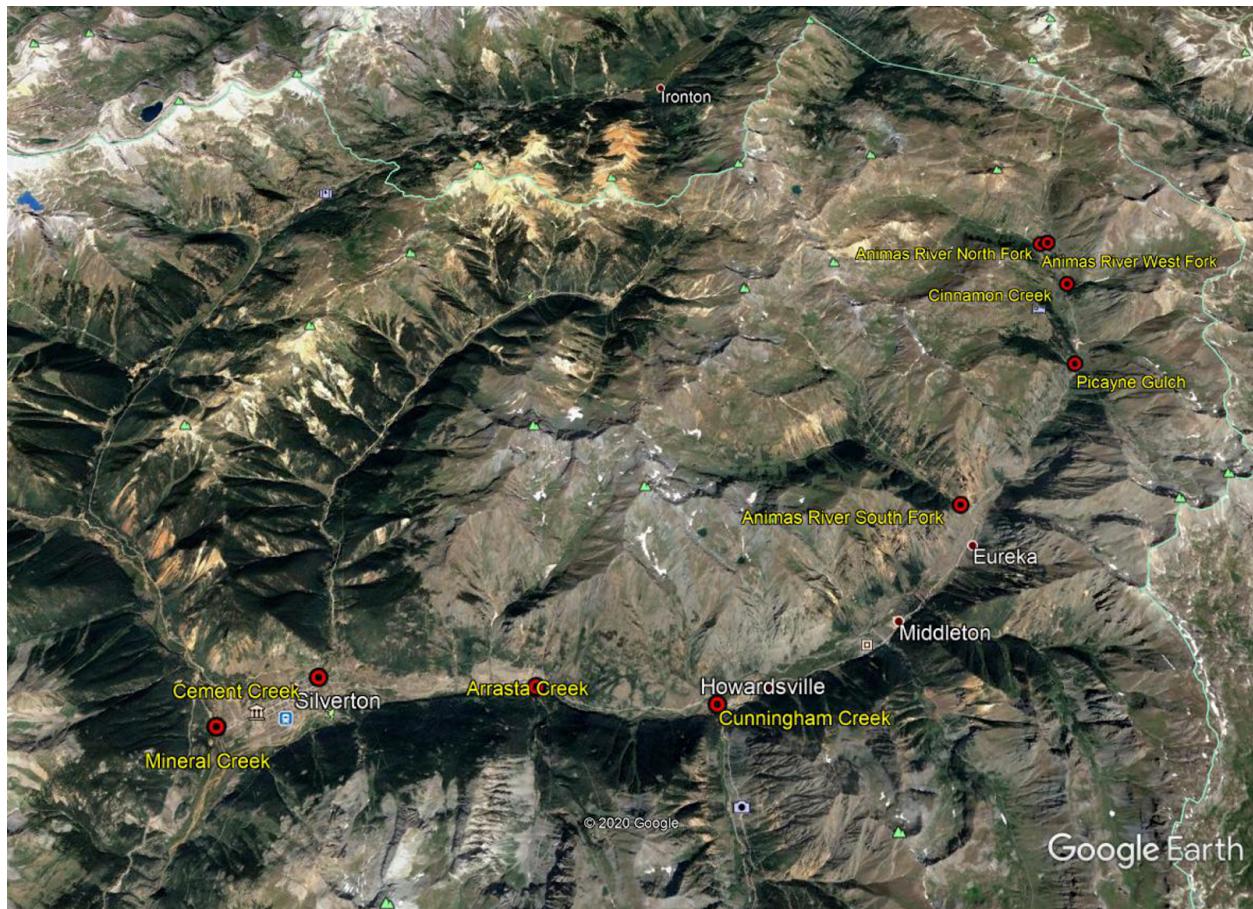
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|                                       |                          |                          |
|---------------------------------------|--------------------------|--------------------------|
| Unnamed Zinc Mine                     | Zinc                     | Gold and Silver          |
| Unnamed Zinc Mine                     | Zinc                     | Copper, Gold, and Lead   |
| Unnamed Zinc Mine                     | Zinc                     | Copper, Gold, and Lead   |
| Unnamed Molybdenum Occurrence         | Molybdenum               | NA                       |
| Unnamed Molybdenum Occurrence         | Molybdenum               | NA                       |
| Brooklyn Silver Mine                  | Copper, Gold, and Lead   | NA                       |
| Lower Browns Gulch Mine               | Zinc                     | Copper, Gold, and Lead   |
| Chattanooga Gold Mine                 | Gold                     | Lead, Silver, and Zinc   |
| Unnamed Gold Mine                     | Gold                     | Copper, Lead, and Silver |
| Unnamed Zinc Group                    | Zinc                     | Copper, Gold, and Lead   |
| Goldfinch Group                       | Molybdenum               | NA                       |
| Goldfinch Mine                        | Molybdenum               | NA                       |
| Imogene Zinc Mine                     | Zinc                     | Gold, Lead, and Silver   |
| Hidden Treasure Molybdenum Occurrence | Molybdenum               | NA                       |
| Upper Mill Creek Mine                 | Gold                     | Lead, Silver, and Zinc   |
| Unnamed Gold Mine                     | Gold                     | Lead, Silver, and Zinc   |
| Silver Crown Mine                     | Lead                     | Silver and Zinc          |
| Unnamed Zinc Mine                     | Zinc                     | Gold and Silver          |
| Unnamed Zinc Mine                     | Zinc                     | Copper, Gold, and Lead   |
| Unnamed Gold Mine                     | Gold                     | Copper                   |
| Unnamed Gold Mine                     | Gold                     | Lead, Silver, and Zinc   |
| Unnamed Zinc Mine                     | Zinc                     | Copper, Gold, and Lead   |
| Mayday Lead Mine                      | Lead, Tungsten, and Zinc | Copper                   |
| Magnet Silver Mine                    | Silver                   | NA                       |

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|                                |          |                          |
|--------------------------------|----------|--------------------------|
| Porphyry Basin Mine            | Gold     | Lead, Silver, and Zinc   |
| Lower Porphyry Gulch Lead Site | Lead     | Gold, Silver, and Zinc   |
| Lower Porphyry Gulch Mine      | Lead     | Silver                   |
| Unnamed Gold Mine              | Gold     | Lead, Silver, and Zinc   |
| Unnamed Gold Mine              | Gold     | NA                       |
| Unnamed Gold Mine              | Gold     | Silver                   |
| Unnamed Gold Group             | Gold     | Lead, Silver, and Zinc   |
| Hoosier Bay Mine               | Tungsten | NA                       |
| U.S. Basin Gold Mine           | Gold     | Silver                   |
| Unnamed Zinc Mine              | Zinc     | Copper, Gold, and Lead   |
| Unnamed Zinc Mine              | Zinc     | Copper, Gold, and Lead   |
| Unnamed Copper Mine            | Copper   | Lead and Zinc            |
| Unnamed Zinc Mine              | Zinc     | Copper, Gold, and Lead   |
| Unnamed Zinc Mine              | Zinc     | Copper, Gold, and Lead   |
| Unnamed Zinc Mine              | Zinc     | Copper, Gold, and Lead   |
| Saint Paul Mine                | Gold     | Copper, Lead, and Silver |
| Unnamed Gold Mine              | Gold     | Lead, Silver, and Zinc   |
| Unnamed Zinc Mine              | Zinc     | Copper, Gold, and Lead   |
| Mineral Basin Silver Site      | Silver   | Copper, Gold, and Lead   |
| Koehler Tunnel                 | Copper   | Lead and Zinc            |

**Appendix III: Maps of the Silverton Caldera**



*The Toxicity of the Lotic Water Bodies In the Silverton Caldera Concerning the Ability To Support  
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**Appendix IV: Photographs From In the Field**

**Animas River West Fork**



*Photos taken by Gavin Tweedie and Evan Tweedie*

*The Toxicity of the Lotic Water Bodies In the Silverton Caldera Concerning the Ability To Support  
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*Animas River North Fork*



*Photos taken by Gavin Tweedie and Evan Tweedie*

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Hexagenia limbata, the Giant Mayfly  
By Evan Tweedie*

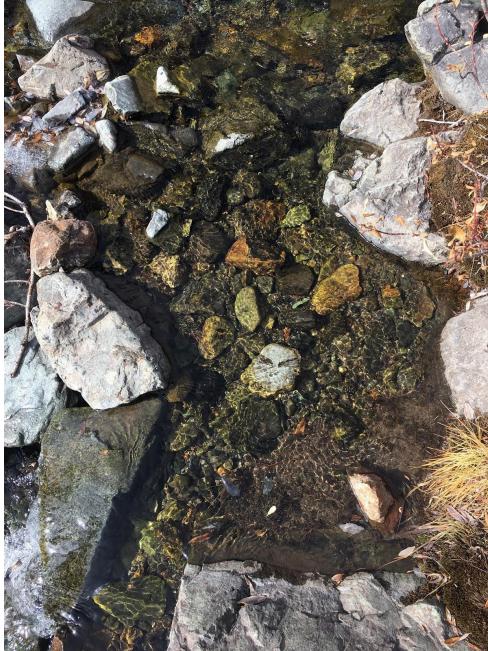
**Cinnamon Creek**



*Photos taken by Gavin Tweedie and Evan Tweedie*

*The Toxicity of the Lotic Water Bodies In the Silverton Caldera Concerning the Ability To Support  
Hexagenia limbata, the Giant Mayfly  
By Evan Tweedie*

**Picayne Gulch**



*Photos taken by Gavin Tweedie and Evan Tweedie*

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***Animas River South Fork***



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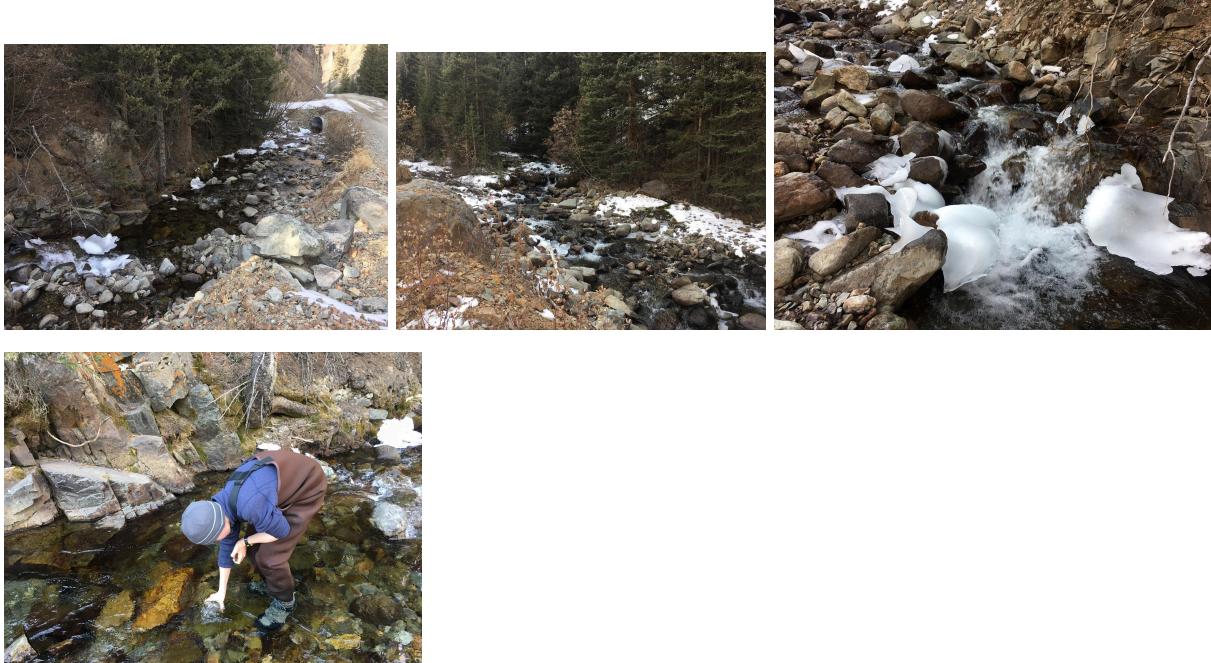
**Cunningham Creek**



*Photos taken by Gavin Tweedie and Evan Tweedie*

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**Arrasta Creek**



*Photos taken by Gavin Tweedie and Evan Tweedie*

*The Toxicity of the Lotic Water Bodies In the Silverton Caldera Concerning the Ability To Support  
Hexagenia limbata, the Giant Mayfly  
By Evan Tweedie*

**Cement Creek**



*Photos taken by Gavin Tweedie and Evan Tweedie*

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Hexagenia limbata, the Giant Mayfly  
By Evan Tweedie*

**Mineral Creek**



*Photos taken by Gavin Tweedie and Evan Tweedie*