

**Geochemistry & Ecology of Red Mat Systems (GERMS)
Teacher Summer Research Program**

Red Layer Microbial Observatory (RLMO)
National Science Foundation
Western Oregon University
Yellowstone National Park



In Yellowstone
Water Chemistry - One of Two Rotating Teams

Your Duties

Set up your work area, paying close attention to waste bottles and efficient labeling
Obtain two 1L bottles of water from the spring source with assistance - label according to...
One 1L analyzed in the field for basic salts; other will be acidified and analyzed for metals at WOU

Site Photography – Shoot At Least 1 High Quality Picture of each of the following...

Water collection in progress, emphasizing the spring features
Please download your images to the computer back at the hotel – and save your disks

Site Parameters (carefully measure and record)

Water temperature at collection site - remember to wipe down probe with alcohol before use
Water pH at collection site

Mat collection team is using the same equipment – check with them.

Chemistry Supplies and Set-Up

Prepare in-field chemistry testing kits (see next table); carry 3-4 of each test/site so we have spares
AccuVac (AV) systems should be carried and sub-divided into extra styrofoam AV containers
Chemical Mix (CM) systems should be properly packed as some compounds are toxic or corrosive
Carry/use plastic container for Hazardous Waste - nitrate ONLY (this must be brought back to WOU)
Non-nitrate liquid can be dumped on site (in bushes away from feature)
Non-nitrate solid is stored in non-hazardous waste designated bottle – dispose at hotel nightly

Following field chemistry, remaining water will be acidified at the hotel via the addition of 5 ml 50% nitric acid per liter sample. Don't forget to do this!

Field Chemical Tests

AccuVac - Your Best Friend

Test Protocol With Water Collected On-Site	Special Notes
Bromine - SAMPLE ACCUVAC AS CHLORINE PROGRAM 6 Fill sample cell with 10 ml sample (BLANK) and ZERO Fill DPD Total Chlorine AccuVac with sample (SAMPLE) - invert Press TIMER ENTER (3 minutes) - READ Sample Pink indicates bromine	Analyze IMMEDIATELY after collection. Known chlorine/derivatives and manganese interfere - all levels. Actual detection limit: 0.03 mg/L.
Total Chlorine PROGRAM 11 Fill sample cell with 10 ml sample (BLANK) and ZERO Fill DPD Total Chlorine AccuVac with sample (SAMPLE) - invert Press TIMER ENTER (3 minutes) - READ Sample Pink indicates chlorine	Analyze IMMEDIATELY after collection. Known bromine/derivatives and manganese interfere - all levels. Actual detection limit: 0.02 mg/L.
Chromium	Analyze IMMEDIATELY after collection.

<p>PROGRAM 14 Fill Chromaver3 with sample (SAMPLE) - invert Press TIMER ENTER (5 minutes) - wait Fill sample cell with 10 ml sample (BLANK) and ZERO READ SAMPLE Purple indicates chromium</p>	<p>Mercury and iron (over 1 mg/L) interferes. Actual detection limit: 0.01 mg/L.</p>
<p><u>Molybdenum</u> PROGRAM 44 Fill sample cell with 10 ml sample (BLANK) Add 4 drops 0.4 CDTA to 40 ml pre-AccuVac beaker sample Fill MolyVer 6 AccuVac with sample - invert Press TIMER ENTER (5 minutes) ZERO BLANK and READ SAMPLE Yellow indicates molybdenum</p>	<p>Analyze IMMEDIATELY after collection. Actual detection limit: 0.2 mg/L. Some metals at high levels (50+ mg/L) can interfere.</p>
<p><u>Nitrate</u> PROGRAM 50 Fill NitraVer AccuVac with sample (SAMPLE) - invert Press TIMER ENTER (1 minute) - MIX SAMPLE ACCUVAC At first timer beep, press ENTER (5 minutes) and wait, no mixing Fill sample cell with 10 ml sample (BLANK) and ZERO READ SAMPLE Amber indicates nitrate</p>	<p>Analyze IMMEDIATELY after collection. All ferric iron, nitrite, and chloride interferes. Actual detection limit: 0.5 mg/L.</p>
<p><u>Nitrite</u> PROGRAM 62 Fill NitriVer AccuVac with sample (SAMPLE) - invert Press TIMER ENTER (15 minutes) Fill sample cell with 10 ml sample (BLANK) and ZERO READ SAMPLE Pink indicates nitrite</p>	<p>Analyze IMMEDIATELY after collection. Watch for precipitation by ions - this will cause interference. Actual detection limit: 0.005 mg/L.</p>
<p><u>Sulfate</u> PROGRAM 92 Fill sample cell with 10 ml (BLANK) Fill Sulfate AccuVac with sample (SAMPLE) - shake 30 seconds Press TIMER ENTER (5 minutes) ZERO BLANK and READ SAMPLE Turbidity indicates sulfate</p>	<p>Analyze IMMEDIATELY after collection. No major interferences. Actual detection limit: 3 mg/L.</p>

Non-AccuVac - Your Good Acquaintance

Test Protocol With Water Collected On-Site	Special Notes
<p><u>Sulfide</u> PROGRAM 93 Fill sample cell with 25 ml (SAMPLE) Fill second cell with 25 ml deionized water (BLANK) Add 1 ml Sulfide 1 Reagent to each - swirl Add 1 ml Sulfide 2 Reagent to each - swirl Press TIMER ENTER (5 minutes) and ZERO BLANK READ SAMPLE Pink indicates sulfide</p>	<p>Analyze IMMEDIATELY after collection. Turbid samples interfere; no other majors interferences. Actual detection limit: 0.01 mg/L. Product contains chromium, disposed of as hazardous waste.</p>

In Yellowstone

Site and Mat Analysis - One of Two Rotating Teams

Your Duties

Set up your work area, paying close attention to clean areas and efficient labeling

Photodocument site - working with water collection team to take pictures

Site Photography – Shoot At Least 1 High Quality Picture of each of the following...

Comprehensive site overview – shows community and nearby regional surroundings

Mat close-up – details area where cores are taken, and preferably shows source

Mat core close-up – well-focused picture of mat core(s), detailing red layer

Please download your images to the computer back at the hotel – and save your disks

Sample Site Overview



Sample Mat Close-Up



Sample Core Close-Up



Site Parameters (carefully measure and record)

Mat temperature where core(s) removed – remember to wipe down probe with alcohol before use

Mat pH where core(s) removed

In the Field Mat Sampling – Target Layer Samples

Your instructor and you will work at the site with different size corers to find the best mat sample for further dissection. Because of topographically complex mat structures, there is no one size fits all.

Mat Dissection

After cores have been removed, carefully place core in sterile Petri dish

Using fresh scalpel blade(s) and flame sterilized forceps, dissect mat into red and green layers

Spend a reasonable amount of time “cleaning” each layer – paring down wrong colors, debris, etc.

Cut final, cleaned mat layers into quarters and place each in labeled, sterile, 1.5 ml microfuge tubes

Label means: white tape with full, legible label (we will assist with designation) covered with clear tape. Write an additional shorthand label on the lid in case the tape comes off during freezing.

Mat Freezing

A dry ice container will be in the van - place your samples immediately in this container

We will put each days' samples in a different plastic freezer bag that has additional labels