KAMIAK BUTTE FIELD SAMPLE ANALYSIS

Carbon & Nitrogen Sequestration

ABSTRACT

Kamiak Butte rests on top of a mountain of quartzite deposited when the region was ocean front property. Loess soils overtopped the butte after glacial-melt flooding scoured the region, and winds blew in the new top-soils. Forests grow on the butte and this report seeks insights to Carbon and Nitrogen sequestration by these tree species.

Hannah Abu-Zahra, Stephanie Kingham, Patrick McCornack, Maria Tooker, Isabella Warren, Jenny Carlson, William E. Schlosser

SoE-300

Kamiak Butte Field Sample Analysis

Hannah Abu-Zahra, SoE-300 Teaching Assistant
Stephanie Kingham, SoE-300 Teaching Assistant
Patrick McCornack, SoE-300 Teaching Assistant
Maria Tooker, SoE-300 Teaching Assistant
Isabella Warren, SoE-300 Teaching Assistant
Jenny Carlson, NSARU Mass Spectrometry Lab Technician
William E. Schlosser, Ph.D.

April 8, 2020

Completed as part of Natural Resource Ecology field data collection and processing events, Spring 2020.

Table of Contents

List of Figures

List of Tables

Acronyms Used

KAMIAK BUTTE FIELD SAMPLE ANALYSIS

The purpose of this paper is to lay out what a mass spectrometry does and the information that can provide information about the amount of carbon and nitrogen being produced on the north and south aspects of Kamiak butte. Other parts of this paper explain the steps that are taken to collecting each sample and preparing them to be placed into mass spectrometry analysis. The last part of this paper explains the results of the data from the mass spectrometry.

Mass Spectrometry

Mass Spectrometry (MS) is an analytical technique that combusts a sample of organic material into gas to measure the mass-to-charge ratio of ions (Sparkman, 2000). This mass spectrometer used for processing samples taken at Kamiak Butte is a coupled system which includes an Elemental Analyzer (EA) and an Isotope Ratio Mass Spectrometer (IRMS). These services and equipment are maintained at Washington State University (WSU) Northwest Sustainable Agroecosystems Research Unit (NSARU) Mass Spectrometry laboratory in Pullman, and were used to process the field samples identified in this report. The first component of this system, the EA, combusts the samples of organic materials and separates them into gas. The second component of this system, the IRMS, is the machine that bends the atoms along their flight path within a vacuum using a strong magnet, allowing detectors in the IRMS to precisely measure the amounts of the different atoms/masses present ,and the amounts of different versions of each atom which are known as isotopes.

Twenty tree core samples were collected to produce accurate Carbon and Nitrogen data. Tree core samples were collected by students during class field trips to Kamiak Butte in two sample areas: North Aspect sites, and South Aspect sites. Each site collection area had trees randomly selected where species and Diameter at Breast Height (DBH) were recorded. All samples were collected as part of the Natural Resources Ecology class of the School of the Environment (SoE) -300 at WSU in Spring 2020, during March.

Samples were labeled, weighed, dehydrated, weighed again (Oven Dry Weight (ODW)) and delivered to the Mass Spectrometry lab at WSU. These ODW samples were then ground into a fine powder. This powder was then enclosed in a tin cover capsule and placed into the first component of the mass spectrometer, the EA. These samples were then combusted, and the gas chemically filtered many times to extract all other components in the gas that were not Carbon or Nitrogen. The Carbon and Nitrogen in the purified gas were then spaced out using a long physical filter so that they could be detected and measured separately per sample. This then led to the second component of the mass spectrometer, the IRMS. The gas was funneled from the EA to the IRMS through hallow wires. Inside the IRMS, the cloud of gas was exposed to strong magnets. These magnets separate the Isotopes by their weight by bending the lighter isotopes closer the magnets as the gas travels through a vacuumed chamber. These separated isotopes were then counted using detectors. The data from these detectors was received as electronic signals which were then compared to known reference materials.

Collecting the samples

Tree core samples were collected from both the north and south aspect of Kamiak butte. The students in each group were tasked with gathering samples, labeling them and following the steps below for how to use the increment bore and successfully obtain both samples. They also used field collection tools listed below in the following section.

Field Tools

Forestry tree increment borers were used to sample tree cores from selected trees at each field plot site. The increment borers are used to manually drill into trees horizontally, entering at DBH on the uphill side of each sampled tree. Additional supplies for this event included a straw to store each sample, pen to label samples, and core storage sealant tape (Figure 1).

Figure 1. *Increment bore device and plastic storage pipets (Abu-Zahra, 2020).*

Steps for collecting the tree core samples

Forest inventory plot centers flags were used to identify the center of each sampled plot. From the plot center the sample crew leader took a compass bearing to determine north, then spun in a northerly direction to identify the first tree within the variable radius plot. The first tree encountered was selected as the increment bore sample tree.

The increment bore device was then assembled. The extractor's threaded end was pushed the into the bark to hang temporarily. Extraction was targeted to enter the sample tree at DBH on the uphill side of the tree. The bill was manually pushed into the tree as it was turned in the clockwise rotation to drill into the tree bole, reaching approximately halfway into the diameter of the tree. When fully entered, the extractor was slowly inserted into the tube in the middle of the handle. The next step was to turn the handle counterclockwise twice until the handle was vertical.

The extractor was then gently pulled to reveal the solid wood core from the sample tree. Students then counted the number of dark rings to determine the age of the tree at breast height. Samples ends were manually removed because this area contains small sections of bark and cambium and this material was not identified for the sample process. This core sample was then placed into the plastic sample receptacles where the label was attached, then sealed at the ends to prevent foreign material entry (Figure 2).

Figure 2. *Collecting the tree core sample (Abu-Zahra, 2020).*

Removal of the increment borer

Increment bore tool removal begins by rotating it counterclockwise until fully removed from the tree. Core sample tube cleaning used a nail to pop the center of the core sample out of the bill and to make sure it does not drop into the straw where the sample is stored. This helps to avoid contamination when taking subsequent samples.

The extractor is then placed in the bill, and the bill into the handle, to lock it up. The samples were then secured for travel in the forest to the next sample site.

Sample Processing

After tree samples were removed from the increment borer and placed into a plastic sample straw, the ends were sealed with tape. The tape is labeled with the group number, date, tree species, and whether the sample was collected from the north or south aspect. These samples are then put in cold storage to prevent them from rotting, or growing mold, before dehydration. The water was removed using the drying oven at the WSU Steffen Center. To accomplish this, each sample was transferred to a labelled coffee filter and weighed. The sample and coffee filter were then placed into the drying for 48 hours at 60°C (Figure 3). After the time had elapsed, they were removed from the oven and weighed again before being moved into labelled water tight sealed bags. The last processing step is to be ground into a fine powder and placed into storage to await mass spectrometry.

Figure 3. *Weighing a green tree core sample (Schlosser W. , 2020)*

Tree Core Results

The samples themselves and the data derived from the mass spectrometry process reveals the net differences in carbon and nitrogen compositions of trees of the north aspect versus the south aspect of Kamiak Butte (Carlson, 2020). These differences are important to understand, as they show variance in ecological character of the two aspects and the growth conditions the various trees are subjected to (Table 1).

Because of the favorable growth conditions of the north aspect for trees and shrubs, vegetation biomass can amass more growth through taller trees with larger tree bole diameters. In contrast, the south aspect vegetation faces hot summertime direct sunlight, and is stressed by less water availability during the late growing periods of July and August. This is combined with shallow quartzite soils with limited water retention capabilities. South aspect tree species include only ponderosa pine with grasses, forbs, and infrequent service berry shrubs. The biomass recycling on this site limits the degradation of nutrients such as nitrogen.

Table 1. EA-IRMS Final Report (Schlosser & Carlson, 2020).

				Total N%			Total C%			C:N Ratio			
Sample Letter	Sample Spp	DBH	ODWg	PSME	LAOX	PIPO	PSME	LAOX	PIPO	PSME	LAOX	PIPO	Aspect
A	PIPO	28.8	1.64			0.131			51.036			388.236:1	North
B	PSME	17.2	1.01	0.105			51.037			485.956:1			North
C	PIPO		0.96			0.102			50.755			496.729:1	South
D	LAOX	19	2.57		0.115			50.255			436.755:1		North
E	PIPO	14.5	1.33			0.139			52.464			378.300:1	South
F	PIPO	26.6	2.25			0.147			51.626			352.180:1	North
G	LAOX		2.12		0.119			49.913			420.923:1		North
H	PIPO	33.3	1.12			0.142			54.174			381.513:1	South
I	PIPO	29	2.27			0.123			56.113			454.549:1	South
J	PSME	28.5	1.48	0.121			50.688			418.713:1			North
K	PIPO		3.13			0.140			59.513			423.808:1	South
L	PIPO		2.04			0.157			55.291			351.245:1	South
M	PIPO	27	1.09			0.192			50.368			262.477:1	South
N	PIPO		1.15			0.140			59.914			428.847:1	South
O	LAOX		1.41		0.121			48.726			402.456:1		North
P	PIPO	14.5	1.6			0.157			50.508			321.446:1	South
Q	PSME	12.5	2.16	0.120			50.321			419.498:1			North
R	LAOX	12.2	2.93		0.118			48.385			410.557:1		North
S	PIPO	10.4	1.8			0.119			54.366			455.773:1	South
T	PIPO	13.0	1.06			0.156			50.060			321.621:1	South
	All Samples	Avg		0.115	0.118	0.142	50.682	49.320	53.553	441.389	417.673	385.902	
		Standard Deviation (σ)		0.009	0.002	0.022	0.358	0.904	3.382	38.598	14.797	65.255	

Where PIPO=*Pinus ponderosa* (ponderosa pine), PSME=*Pseudotsuga menziesii* (Douglas-fir), and LAOX=*Larix occidentalis* (western larch).

While it may make sense to hypothesize that the north facing aspect would have higher concentrations of carbon and nitrogen due to more ideal growth conditions, the results found through analyzing the samples from the two aspects are remarkably similar. Each species and each aspect of samples carried approximately 0.133% Nitrogen (σ=0.022), and 52.276% Carbon (σ=52.276) (Table 2).

Table 2. Average sample statistics, all species and all aspects (Schlosser & Carlson, 2020).

	Total N%	Total C%	C:N Ratio
Average all samples	0.133	52.276	400.579
Standard Deviation (σ)	0.022	3.275	57.903
Number of samples	20	20	20

While the ratios of carbon to nitrogen were remarkably similar, the largest difference is that the north facing aspect had greater amounts of biomass than the south facing aspect, since it had more ideal growth conditions. To use the data obtained from mass spectrometry analysis, data compared concentrations of nitrogen and carbon between core samples from both aspects. While comparing, note the biomass associated with each sample/from each aspect (Table 1). Create ratios of carbon to nitrogen to compare the results of the two aspects. To summarize (Table 2), note that the ratio is similar across both aspects; the difference comes from differences in biomass.

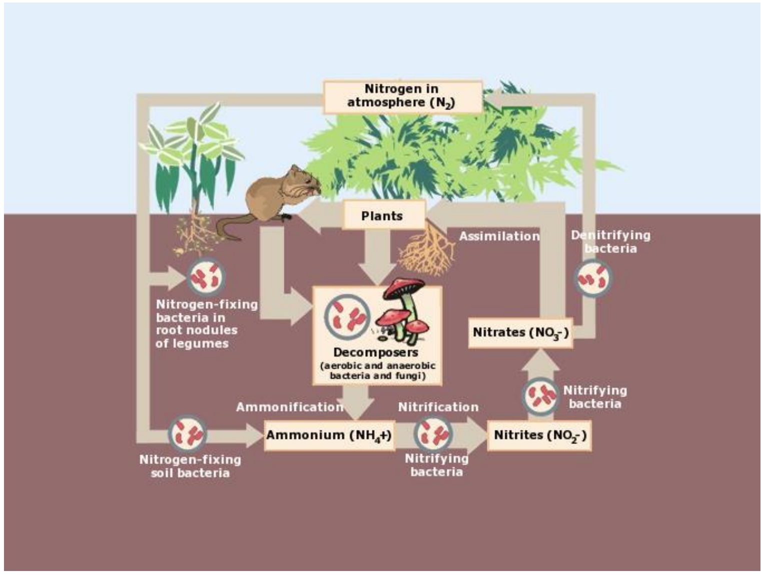
The differences are witnessed in the sizes of the trees on each aspect.

Conclusion

Nitrogen Sequestration

Nitrogen is a very important element that plants use in order to thrive and survive. Nitrogen is filtered through the environment via the nitrogen cycle (Figure 4). A part of this cycle is decomposers breaking down organisms that have died and turning it into ammonia. Ammonia is then turned into nitrates and nitrites by nitrifying bacteria. Plants take up nitrates by assimilation (The Editors of Encyclopaedia Britannica, 2020). Within the north side of Kamiak Butte, more nitrogen is available for plants to take up. This is because there is a lot more plant life on the north side and when a plant drops dead leaves, the decomposers act upon it, turning it to ammonium. The ammonium will go through the nitrogen cycle until it becomes nitrate, which is the compound that plants can assimilate. Nitrogen is needed for the plant because it helps with chloroplast development. It is also necessary for the functioning of photosynthesis and other enzyme biochemical processes (Schlosser, Nitrogen and Carbon and it's Affects on Kamiak Butte, 2020). The trees on the north facing aspect have higher net productivity than the trees on the south aspect because of this. The humus on the north facing aspect also prevents leaching and the loss of nitrogen through the soils, which can be prevalent on Kamiak Butte because of it being set on fractured quartzite. The south facing aspect does not have as many plants on it. Having less dead plant matter reduces the amount of nitrogen it can assimilate. The nitrogen that can be taken up is used towards processes like chloroplast production. Chloroplasts are used by the plants to sequester atmospheric Carbon. Since not as much nitrogen is available, there is not as much chloroplast production, and thereby not as much Carbon can be sequestered. This is one of the reason sets why trees on the south facing aspect are stunted in growth in comparison to the north facing aspect.

Figure 4. *Nitrogen Cycle (EarthHow, 2019)*



Carbon Sequestration

Carbon is an element used by plants that allows them to do certain types of processes. Carbon is sequestered through photosynthesis, a process that converts carbon dioxide and water into glucose and oxygen (Kaufman & Franz, 1996). Atmospheric Carbon is readily available for both aspects of Kamiak Butte and the assumption could be made that there would be equal amounts of carbon on both aspects. The amount of nitrogen and available water on the north and south aspects of Kamiak Butte differ. Precipitation timing and temperature are critically important to dissolve nitrogen as plant roots move it along the xylem layer of tree cambium and make it available to plant growth.

Northern aspect trees develop more chloroplasts which absorb sunlight and uses it, together with water and carbon dioxide, to produce sugars that the plants use as food. With there being more chloroplasts developed on the northern aspect, there is more carbon uptake (Schlosser, Nitrogen and Carbon and it's Affects on Kamiak Butte, 2020). The amount of carbon uptake will be a factor in the overall productivity of the plants. Just like with nitrogen, it can be seen with the naked eye that carbon is used more on the northern aspect creating larger trees taking advantage of their favorably restrictive environmental conditions. The northern trees have thicker trunks, more branches and leaves than the southern aspect trees.

Works Cited

Carlson, J. (2020, March 5). Mass Spectrometry. (I. Warren, Interviewer) Pullman, Wa.

EarthHow. (2019, December 16). *What Are the 4 Steps of Nitrogen Cycle?* Retrieved from EarthHow: https://earthhow.com/nitrogen-cycle/

Kaufman, D., & Franz, C. (1996). *Biosphere 2000: protecting our global environment.* Kendall/Hunt Pub. Co. doi:ISBN 978-0-7872-0460-0.

Schlosser, W. (2020, March 22). Nitrogen and Carbon and it's Affects on Kamiak Butte. (S. Kingham, Interviewer) Pullman, Wa.

Schlosser, W. (2020). Weighing tree core dehydrated samples. *Mass Spec. Report.* Washington State University, Pullman, WA.

Schlosser, W., & Carlson, J. (2020). *Mass Spectrometry Results, Kamiak Butte, SP2020.* WSU, SoE. Pullman: Northwest Sustainable Agroecosystems Research Unit.

Sparkman, O. (2000). *Mass spectrometry desk reference.* P{ittsburg: Global View Pub. doi:978-0-9660813-2-9

The Editors of Encyclopaedia Britannica. (2020, February 20). *Nitrogen cycle*. Retrieved from Britannica: https://www.britannica.com/science/nitrogen-cycle