Name: ____

Using Mass Spectrometry to Characterize Complex Organic Mixtures

Introduction

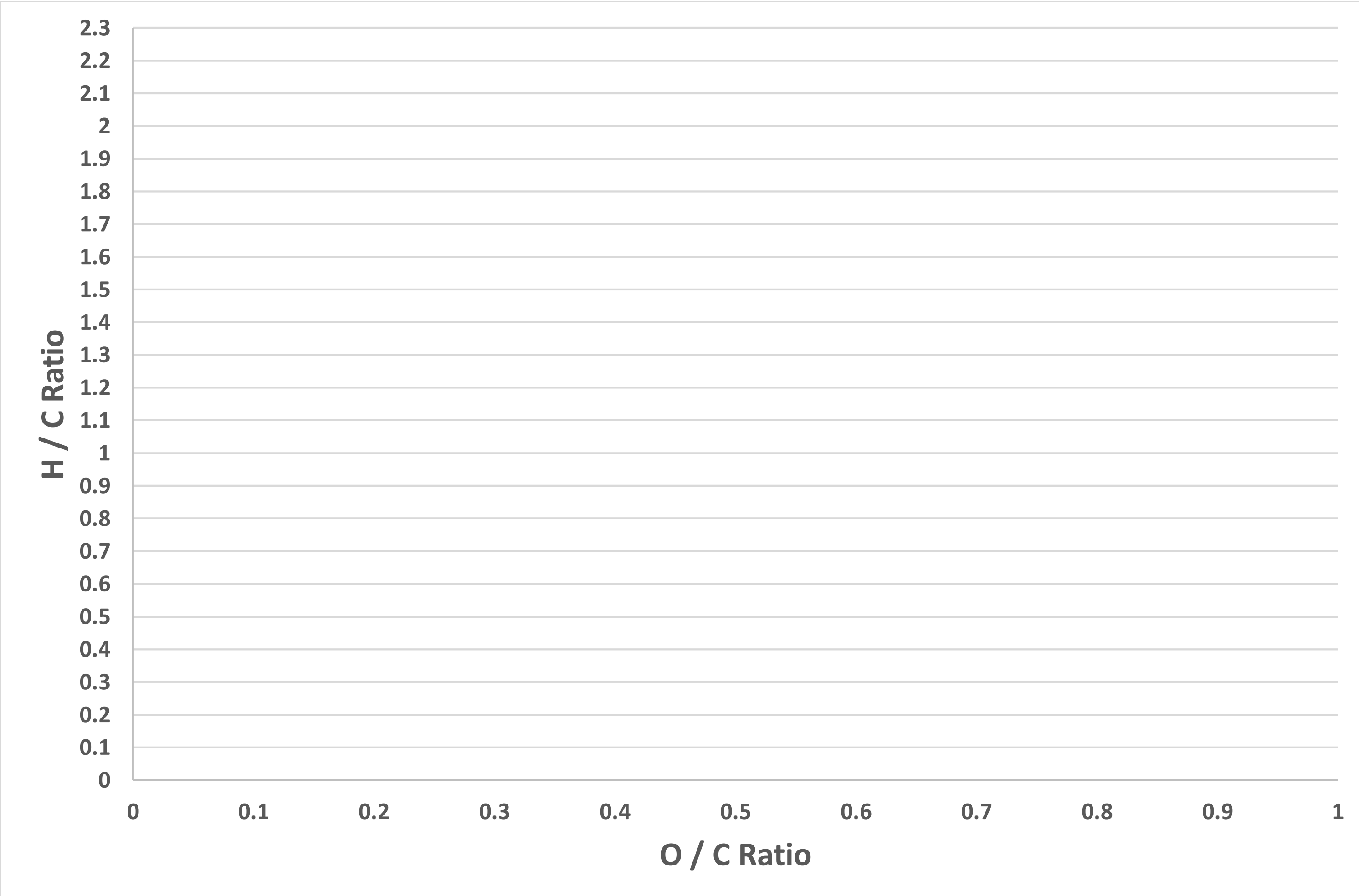
Scientists use high resolution mass spectrometry to characterize the composition of complex organic mixtures found in nature. Molecular weight, hydrogen/carbon (H/C) ratio, and oxygen-carbon (O/C) ratio can be used to determine compound class. Van Krevelen diagrams, which plot H/C ratio as a function of O/C ratio, are very useful in graphically depicting the different components of a mixture. In this series of activities, you will create Van Krevelen diagrams depicting different classes of compounds and learn to recognize patterns in these diagrams. You will then use your knowledge to characterize the organic composition of a sample of stream water.

ACTIVITY 1 – Introduction to Van Krevelen Diagrams

1. Complete the below table for the compounds listed.

Compound	Structural Drawing	Molecular Formula	H / C Ratio	O / C Ratio	Aromatic or Aliphatic?
Hexane					
Toluene					
Glucose					
Acetic Acid					
Benzene					
Pyridine					
1,3-Butadiene					
Benzoic Acid					

2. Create a Van Krevelen diagram for the compounds on the previous page. Plot **aromatic** compounds with an **X** and **aliphatic** compounds with a **dot**.



3. On your diagram, attempt to draw a line to separate aromatic and aliphatic compounds. What conclusion(s) can you draw regarding the general location of aromatic vs. aliphatic compounds on a Van Krevelen diagram?

4. Next to each data point on your diagram, list the number of carbon-oxygen bonds the compound contains. A carbon-oxygen double bond (carbonyl functional group) counts as two carbon-oxygen bonds. What conclusion(s) can you draw regarding compound oxidation and location on a Van Krevelen diagram?

ACTIVITY 2 – Van Krevelen Diagrams and Compound Class

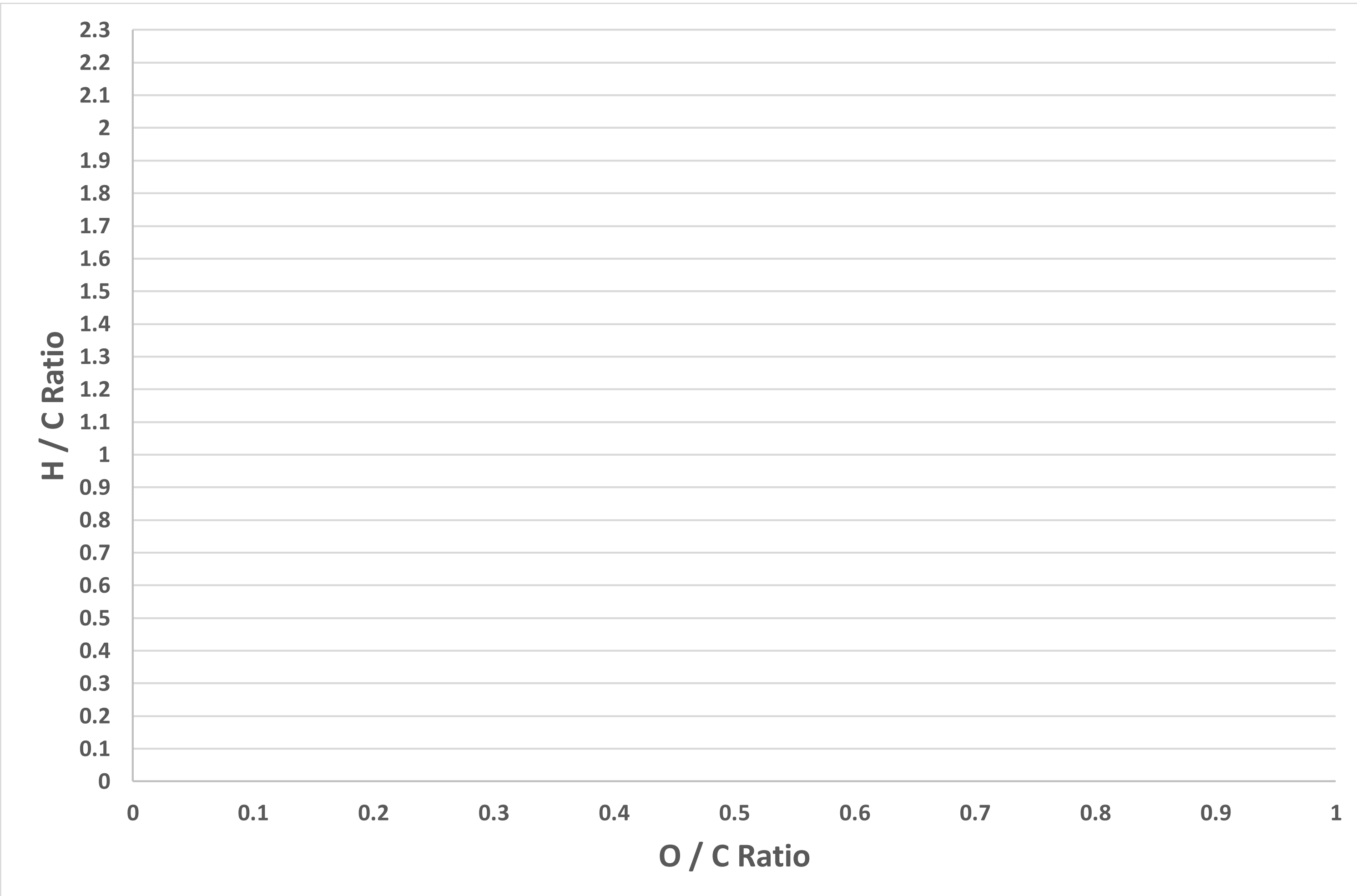
1. Organic mixtures present in nature often contain a wide variety of compound classes. Using resources from the internet, briefly define the below organic compound classes.

   - **Unsaturated Hydrocarbon**
   - **Condensed Aromatic**
   - **Protein**
   - **Lipid**
   - **Carbohydrate**
   - **Tannin**
   - **Lignin**
   - **Amino Sugar**

2. Complete the below table for the compounds listed.

Compound	Compound Class	Molecular Formula	H / C Ratio	O / C Ratio
Cyclohexene	Unsaturated Hydrocarbon	C_6H_{10}		
2-Methyl-2-Butene	Unsaturated Hydrocarbon	C_5H_{10}		
1,3,5-Hexatriene	Unsaturated Hydrocarbon	C_6H_8		
Anthracene	Condensed Aromatic	$C_{14}H_{10}$		
Coronene	Condensed Aromatic	$C_{24}H_{12}$		
Pyrene	Condensed Aromatic	$C_{16}H_{10}$		
Hemoglobin	Protein	$C_{2952}H_{4664}O_{832}N_{812}S_8Fe_4$		
Collagen	Protein	$C_{65}H_{102}N_{18}O_{21}$		
Keratin	Protein	$C_{28}H_{48}N_2O_{32}S_4$		
Tristearin	Lipid	$C_{57}H_{110}O_6$		
Linolein	Lipid	$C_{57}H_{98}O_6$		
Triolein	Lipid	$C_{57}H_{104}O_6$		
Sucrose	Carbohydrate	$C_{12}H_{22}O_{11}$		
Isomaltulose	Carbohydrate	$C_{12}H_{22}O_{11}$		
Glucose	Carbohydrate	$C_6H_{12}O_6$		
Tannic Acid	Tannin	$C_{76}H_{52}O_{46}$		
Castalagin	Tannin	$C_{41}H_{26}O_{26}$		
Gallic Acid	Tannin	$C_7H_6O_5$		
Hardwood Lignin A	Lignin	$C_{81}H_{92}O_{28}$		
Hardwood Lignin B	Lignin	$C_{31}H_{34}O_{11}$		
Hardwood Lignin C	Lignin	$C_{278}H_{407}O_{94}$		
Daunosamine	Amino Sugar	$C_6H_{13}NO_3$		
N-acetylglucoseamine	Amino Sugar	$C_8H_{15}NO_6$		
Galactosamine	Amino Sugar	$C_6H_{13}NO_5$		

3. Create a Van Krevelen diagram for the compounds on the previous page. Use the below key to denote different compound classes on the diagram.
   - **P = Protein**
   - **T = Tannin**
   - **AS = Amino Sugar**
   - **CA = Condensed Aromatic**
   - **LP = Lipid**
   - **LI = Lignin**
   - **CB = Carbohydrate**
   - **UH = Unsaturated Hydrocarbon**



4. On your diagram, draw rectangles around the data for each compound class to create general Van Krevelen compound class regions. Label each of these regions with the associated compound class.

5. On your diagram, draw a line to separate aromatic and aliphatic compounds. This line should resemble that which you created for Activity 1.

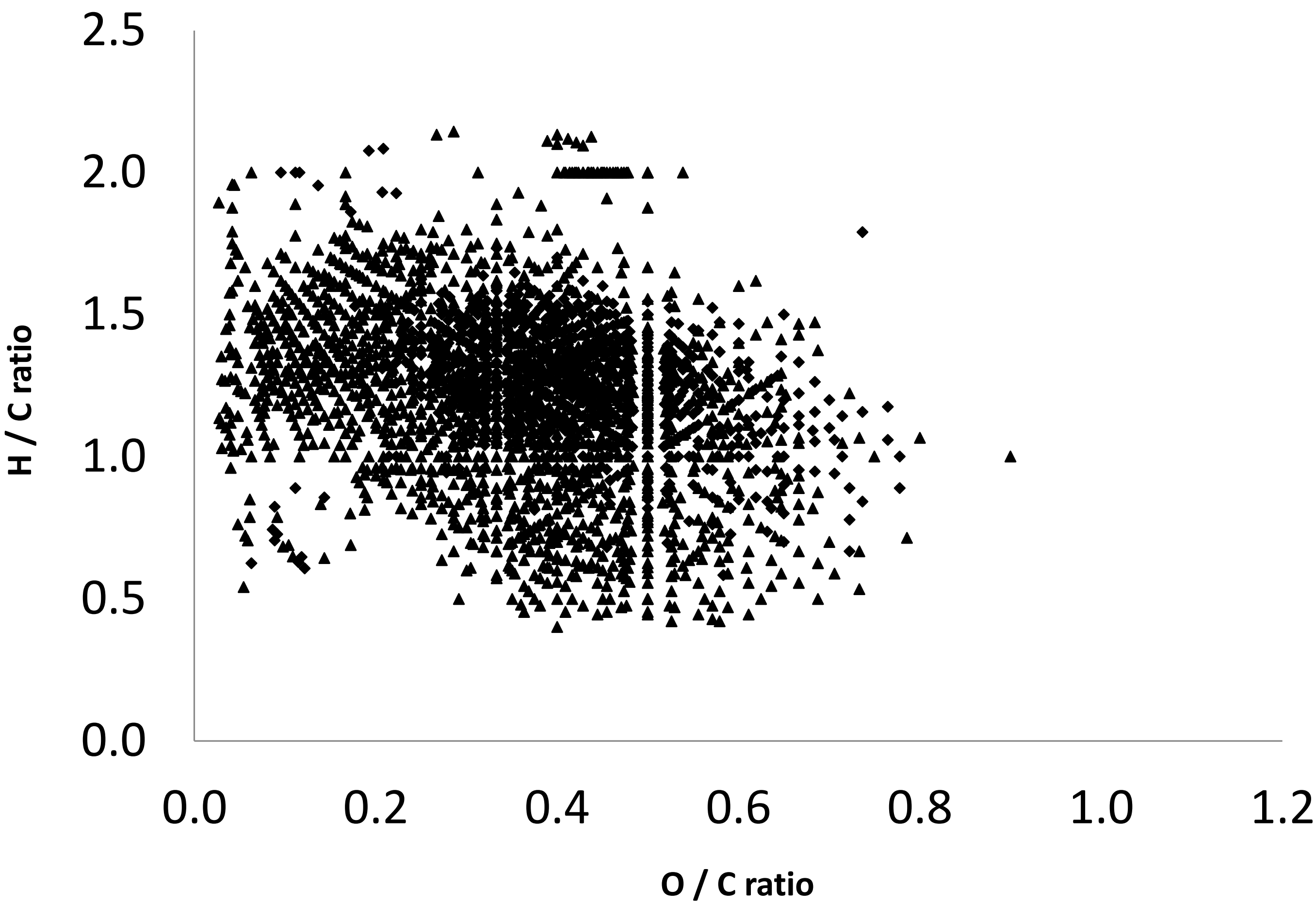
   a. List one compound class that falls entirely below the line (more aromatic). Explain this trend, given the nature of this compound class.

   b. List one compound class that falls entirely above the line (more aliphatic). Explain this trend, given the nature of this compound class.

6. Which compound class is the most oxidized? Explain how you know, using both Van Krevelen diagram data and your knowledge of the nature of the compound class.

ACTIVITY 3 – Characterization of Stream Water Sample

Below is a Van Krevelen diagram for a sample of water from a stream in northern Alaska.



1. What conclusions can you draw regarding the aromatic vs. aliphatic organic content in this water sample?

2. What compound class is most prevalent in this water sample? Given the nature of this compound class (chemical structure, role in nature), attempt to explain why.

3. Which compound classes are least prevalent in this water sample? Given the nature of these compound classes (chemical structure, role in nature), attempt to explain why.

Name: ____

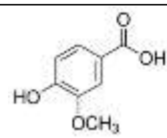
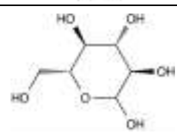
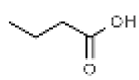
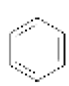
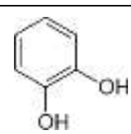
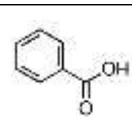
Using Mass Spectrometry to Characterize Complex Organic Mixtures

Introduction

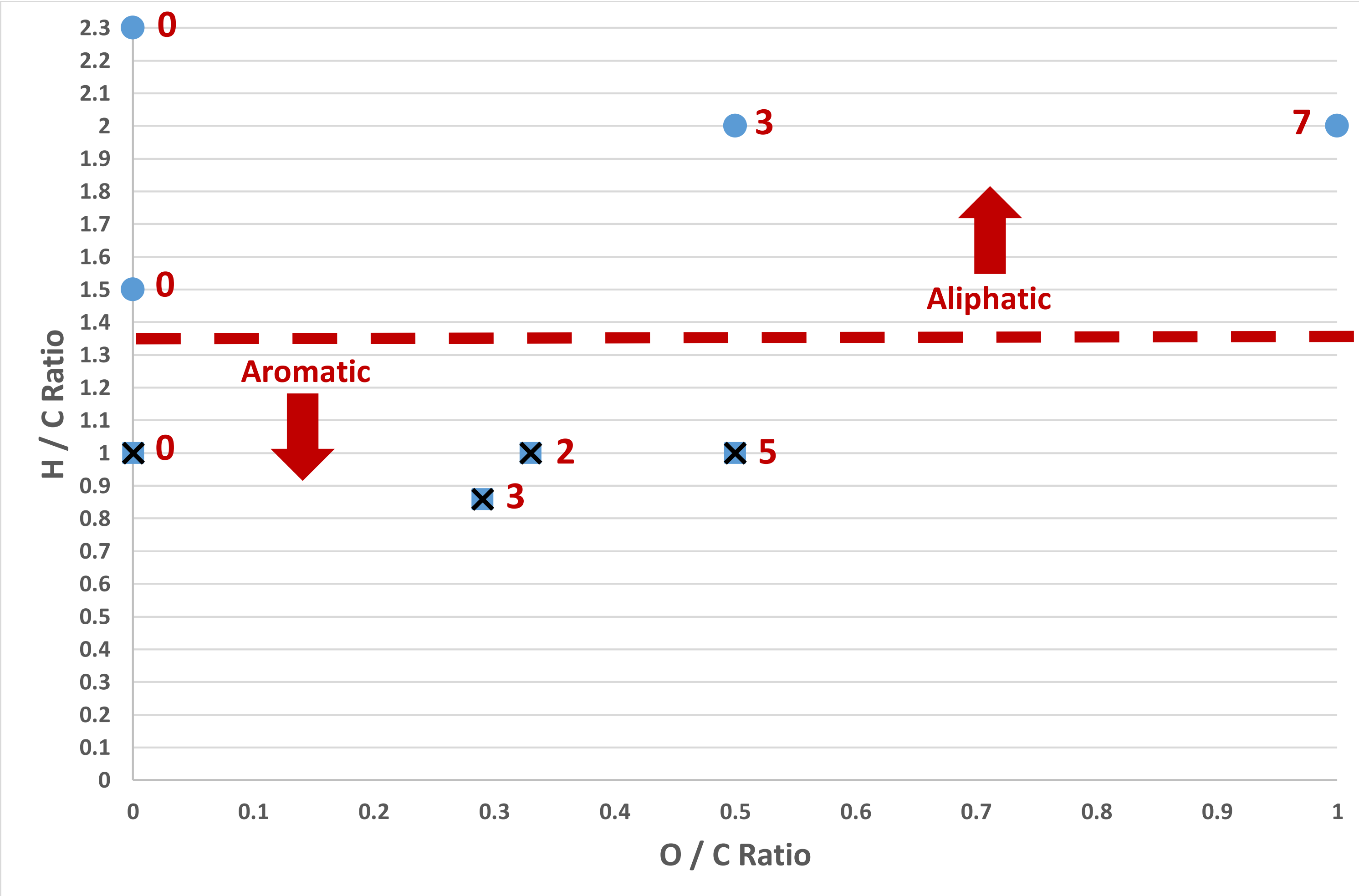
Scientists use high resolution mass spectrometry to characterize the composition of complex organic mixtures found in nature. Molecular weight, hydrogen/carbon (H/C) ratio, and oxygen-carbon (O/C) ratio can be used to determine compound class. Van Krevelen diagrams, which plot H/C ratio as a function of O/C ratio, are very useful in graphically depicting the different components in a mixture. In this series of activities, you will create Van Krevelen diagrams depicting different classes of compounds and learn to recognize patterns in these diagrams. You will then use your knowledge to characterize the organic composition of a sample of stream water.

ACTIVITY 1 – Introduction to Van Krevelen Diagrams

1. Complete the below table for the compounds listed.

Compound	Structural Drawing	Molecular Formula	H / C Ratio	O / C Ratio	Aromatic or Aliphatic?
Hexane		C_6H_{14}	2.3	0.00	Aliphatic
Vanillic Acid		$C_8H_8O_4$	1.0	0.50	Aromatic
Glucose		$C_6H_{12}O_6$	2.0	1.0	Aliphatic
Butyric Acid		$C_4H_8O_2$	2.0	0.50	Aliphatic
Benzene		C_6H_6	1.0	0.00	Aromatic
Catechol		$C_6H_6O_2$	1.0	0.33	Aromatic
1,3-Butadiene		C_4H_6	1.5	0.00	Aliphatic
Benzoic Acid		$C_7H_6O_2$	0.86	0.29	Aromatic

2. Create a Van Krevelen diagram for the compounds on the previous page. Plot **aromatic** compounds with an **X** and **aliphatic** compounds with a **dot**.



3. On your diagram, attempt to draw a line to separate aromatic and aliphatic compounds. What conclusion(s) can you draw regarding the general location of aromatic vs. aliphatic compounds on a Van Krevelen diagram?

   Aromatic compounds are located in the lower half of the Van Krevelen diagram (lower H/C ratio). Aliphatic compounds are location in the upper half (higher H/C ratio).

4. Next to each data point on your diagram, list the number of carbon-oxygen bonds the compound contains. A carbon-oxygen double bond (carbonyl functional group) counts as two carbon-oxygen bonds. What conclusion(s) can you draw regarding compound oxidation and location on a Van Krevelen diagram?

   More oxidized compounds are located further to the right on the Van Krevelen diagram (higher O/C ratio)

ACTIVITY 2 – Van Krevelen Diagrams and Compound Class

1. Organic mixtures present in nature often contain a wide variety of compound classes. Using resources from the internet, briefly define the below organic compound classes.

   - **Unsaturated Hydrocarbon**
     Hydrocarbons that contain pi bonds and/or rings

   - **Condensed Aromatics**
     Compounds that contain fused aromatic rings

   - **Protein**
     Polymer chains of amino acids linked together by peptide bonds

   - **Lipid**
     Fatty acids or their derivatives

   - **Carbohydrate**
     Sugars, starch, or cellulose

   - **Tannin**
     Complex biomolecules present in plant tissue that serve to bind and precipitate proteins. They play a role in plant defense against predation (as they give tissue a characteristic astringency) and are also involved in control of plant growth.

   - **Lignin**
     A class of organic polymers present in the structural tissue of plants. These polymers are extremely abundant in terrestrial plant material and very resistant to biodegradation.

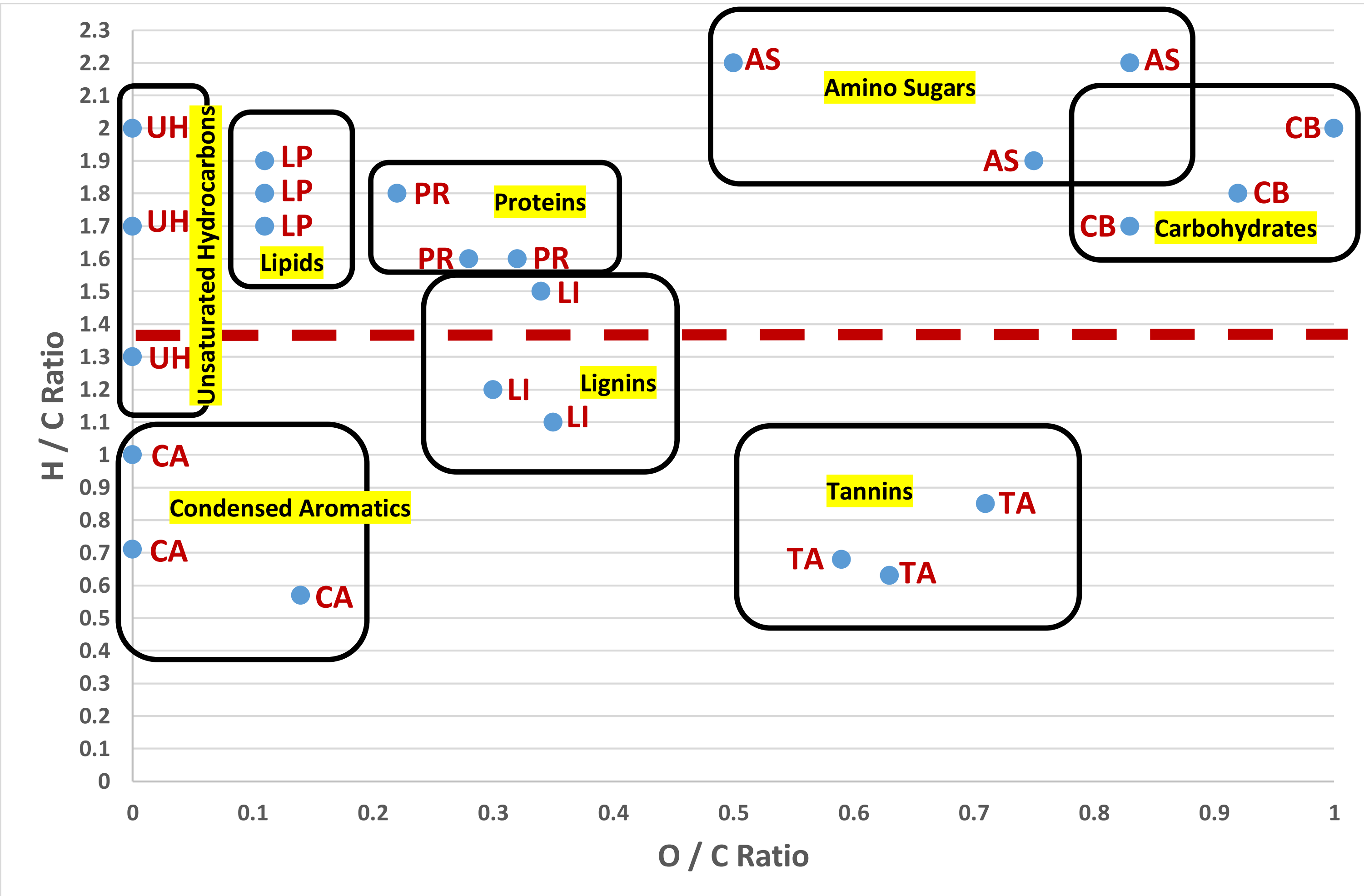
   - **Amino Sugar**
     Sugars in which one of the hydroxyl (-OH) groups has been replaced by an amine group ($-NH_2$, -NHR, $-NR_2$)

2. Complete the below table for the compounds listed.

Compound	Compound Class	Molecular Formula	H / C Ratio	O / C Ratio
Cyclohexene	Unsaturated Hydrocarbon	C_6H_{10}	1.7	0.00
2-Methyl-2-Butene	Unsaturated Hydrocarbon	C_5H_{10}	2.0	0.00
1,3,5-Hexatriene	Unsaturated Hydrocarbon	C_6H_8	1.3	0.00
Anthracene	Condensed Aromatic	$C_{14}H_{10}$	0.71	0.00
Pyridine	Condensed Aromatic	C_5H_5N	1.0	0.00
Anthraquinone	Condensed Aromatic	$C_{14}H_8O_2$	0.57	0.14
Hemoglobin	Protein	$C_{2952}H_{4664}O_{832}N_{812}S_8Fe_4$	1.6	0.28
Collagen	Protein	$C_{65}H_{102}N_{18}O_{21}$	1.6	0.32
Elastin	Protein	$C_{27}H_{48}N_6O_6$	1.8	0.22
Tristearin	Lipid	$C_{57}H_{110}O_6$	1.9	0.11
Linolein	Lipid	$C_{57}H_{98}O_6$	1.7	0.11
Triolein	Lipid	$C_{57}H_{104}O_6$	1.8	0.11
Sucrose	Carbohydrate	$C_{12}H_{22}O_{11}$	1.8	0.92
Starch	Carbohydrate	$C_6H_{10}O_5$	1.7	0.83
Glucose	Carbohydrate	$C_6H_{12}O_6$	2.0	1.0
Tannic Acid	Tannin	$C_{76}H_{52}O_{46}$	0.68	0.59
Castalagin	Tannin	$C_{41}H_{26}O_{26}$	0.63	0.63
Gallic Acid	Tannin	$C_7H_6O_5$	0.85	0.71
Hardwood Lignin A	Lignin	$C_{60}H_{72}O_{18}$	1.2	0.30
Hardwood Lignin B	Lignin	$C_{31}H_{34}O_{11}$	1.1	0.35
Hardwood Lignin C	Lignin	$C_{278}H_{407}O_{94}$	1.5	0.34
Daunosamine	Amino Sugar	$C_6H_{13}NO_3$	2.2	0.50
N-acetylglucoseamine	Amino Sugar	$C_8H_{15}NO_6$	1.9	0.75
Galactosamine	Amino Sugar	$C_6H_{13}NO_5$	2.2	0.83

3. Create a Van Krevelen diagram for the compounds on the previous page. Use the below key to denote different compound classes on the diagram.

- **PR = Protein**
- **TA = Tannin**
- **AS = Amino Sugar**
- **CA = Condensed Aromatics**
- **LP = Lipid**
- **LI = Lignin**
- **CB = Carbohydrate**
- **UH = Unsaturated Hydrocarbon**



4. On your diagram, draw rectangles around the data for each compound class to create general Van Krevelen compound class regions. Label each of these regions with the associated compound class.

5. On your diagram, draw a line to separate aromatic and aliphatic compounds. This line should resemble that which you created for Activity 1.

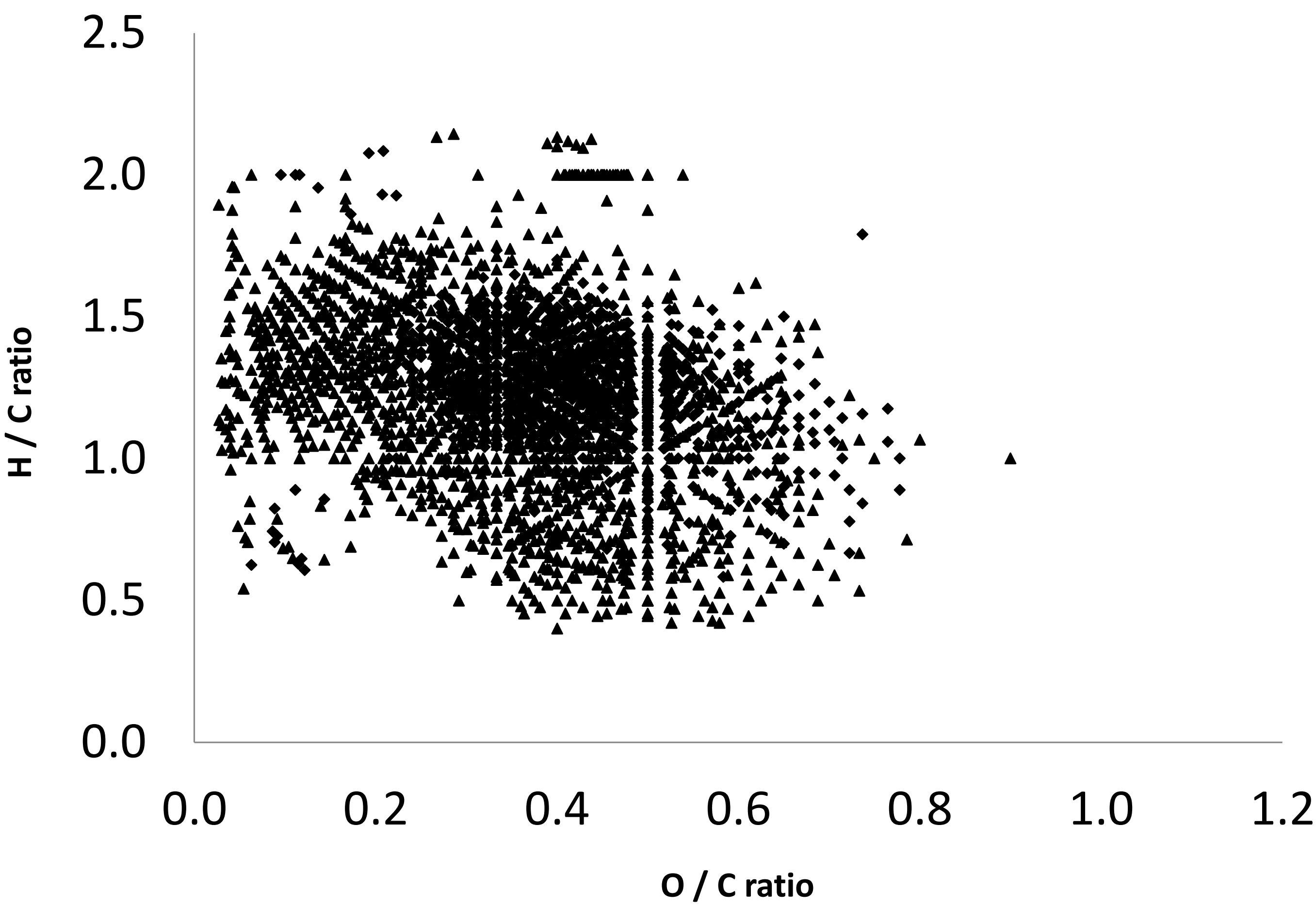
    a. List one compound class that falls entirely below the line (more aromatic). Explain this trend, given the nature of this compound class. Condensed aromatics. They consist of fused aromatic rings.

    b. List one compound class that falls entirely above the line (more aliphatic). Explain this trend, given the nature of this compound class. Lipids. The main categories of lipids are fatty acids, glycerolipids, glycerophospholipids, sphingolipids, sterols, prenols, saccrolipids, and polyketides. None of these compounds contain aromatic rings.

6. Which compound class is the most oxidized? Explain how you know, using both Van Krevelen diagram data and your knowledge of the nature of the compound class. Carbohydrates. From the Van Krevelen data, it is clear that these compounds have very high O/C ratios. As they are hydrates of carbon, most (if not all) carbons in the molecule are bonded to at least one oxygen.

ACTIVITY 3 – Characterization of Stream Water Sample

Below is a Van Krevelen diagram for a sample of water from a stream in northern Alaska.



1. What conclusions can you draw regarding the aromatic vs. aliphatic organic content in this water sample?

   Simliar amount of aromatic and aliphatic compounds are present in the sample.

2. Which compound class is most prevalent in this water sample? Given the nature of this compound classes (chemical structure, role in nature), attempt to explain why.

   Lignins are most prevalent in this water sample. The comonality of these polymers in stream water makes sense, as they are extremely abundant in terrestrial plant material and very resistant to biodegradation.

3. Which compound classes are least prevalent in this water sample? Given the nature of these compound classes (chemical structure, role in nature), attempt to explain why.

   Carbohydrates and amino sugars are least prevalent in this water sample. In streams, these compounds classes serve as imporant nutrient sources for microorganisms. Carbohydrate and amino sugar content in steam water varies drastically based on the microbiological makeup of the stream and time of year.