

THREE NOVEL SCIENCE ACTIVITIES RELATING TO THE STRUCTURE OF THE ATOM,  
BIOINFORMATICS, AND THE DENATURATION OF PROTEIN

by

ROBERT DUERST

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Advisor's Signature

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## Introduction

This paper describes three novel activities that were designed to teach difficult scientific concepts to a wide age range of students (7<sup>th</sup> through 11<sup>th</sup> grade). The subject of the three activities include the structure of the atom, bioinformatics, and protein denaturation. Each section within the paper includes background information and material lists necessary for the activity, in addition to a procedure and reflection.

The two models of learning used to analyze the activities were Bloom's Taxonomy and the Constructivist Theory. In Bloom's Taxonomy, there are six levels in the Cognitive domain: Knowledge, Comprehension, Application, Analysis, Synthesis, and Evaluation (in order from lowest to highest). As a student progresses from the lower levels to the higher levels, their comprehension and mastery of the subject increases. According to Constructivism, a learner needs to be active in the learning process so that they can give meaning to their experiences. This happens when the learner reconciles their experiences to something they previously held to be true.

## Atomic Duck Duck Goose

### I.) Description of Activity

The goal of the activity is to give 5<sup>th</sup> through 7<sup>th</sup> grade students a hands-on way to conceptualize the structure of the atom. After going through this activity students should be able convey that protons and neutrons are in the center of the atom in a region known as the nucleus, that electrons move around the outer regions of the atom in an area called the electron cloud, and that the electron cloud makes up the majority of the atom's volume.

The specific advantage this activity offers younger students is that it will let them use their whole bodies while learning about atomic structure whereas most other activities are pencil and paper or will at best involve making a drawing/model.

#### Materials Needed:

- Blank sheets of paper (two per student)
- Thumb tacks
- Pencil with a dull lead
- Periodic table
- A large space for students to run around (ideally a 400 meter track).

### **Procedure for Part 1 (Classroom Portion):**

1. Have a discussion with students about what matter is made of.
2. Introduce the concept of the “atom” to students.
3. Have students break into groups to brainstorm what an atom is (they may write ideas on a sheet of paper).
4. Write student ideas on board.
5. Play this YouTube video relating to the structure of the atom.  
(<http://www.youtube.com/watch?v=IP57gEWcisY>)
6. As a class, discuss which concepts they had correct, and correct any misconceptions they have had.

Teaching Note: A misconception that students often have is that electrons orbit around the nucleus the way that planets orbit around the Sun. This is incorrect, as electrons will change direction at random as they are moving around the nucleus and have a specific probability of being in a certain space known as the electron cloud. (Krebs, 1999)

7. On a sheet of paper have students draw a correct model of the atom and label the parts.

### **Procedure for Part 2 (Sub atomic Structure):**

1. Number the students off by three and have them write their number on their drawing of the atom.
2. Have students bring their drawing to a large space where they can run around.
3. Assign each group a subatomic particle (proton, neutron, or electron).
4. Ask students where each particle goes.
5. Starting with Helium, have a “proton” and “neutron” go to the nucleus and then have an “electron” move around nucleus at random.
  - a. An important point to emphasize here is that it is the number of protons that determines what the element is. To do this, add multiple neutrons and electrons, and every time you add a neutron or electron, check with the students to make sure they still understand you are working with a Helium atom.
  - b. It is not recommend that you discuss ions with students at this point, but if they ask you should be prepared to mention how the charge changes when electrons are added or subtracted.
6. Direct the electron to move at random around the nucleus.
7. Work through the elements Li, and Be.
8. Design the maximum sized atom possible based on the number of students you have to work with (this will usually be Carbon, Nitrogen, or Oxygen).

### **Procedure for Part 3 (Relative size of an atom)**

1. Bring the students to a 400-yard track and give each student a thumbtack.

2. Explain that the very tip of the thumbtack represents an electron. It is so small that you can't see it with your naked eye.
3. Show the students a dull pencil. Explain that the nucleus (made up of protons and neutrons) is represented by the tip of the pencil.
4. Push the pencil down into the ground in the middle of the track so that the point is facing upwards.
5. Ask the students how far away they think the electrons are supposed to be if the nucleus is that big.
6. Explain that if the nucleus is as big as a pencil point, the electrons should actually be 100 meters away, but that you are going to use the track as a rough approximation.
7. Explain to students that electrons usually stay a specific distance from the nucleus.
8. Divide your students up and put them into the different lanes of the track at different locations around the track.
9. Let the students run around at random (Changing direction as they please) as long as they stay in the same lane of the track.
10. End of activity.

## II). Results

The positive outcomes of this activity were that my students enjoyed themselves and were able to learn about where in the atom each of the subatomic particles are located. There was a very high retention rate among the students based on anecdotal evidence and high quiz scores compared to another class period which did not experience the activity. The quiz consisted of a generic atom where students needed to label the parts and identify which element what represented. A statistical treatment was not done because the experimental group consisted of sixteen 7<sup>th</sup> grade students and the control group consisted primarily of 11<sup>th</sup> grade students. A negative outcome of the implementation of this activity was that it took three class periods and that going outdoors is weather dependent (coordinating with the physical education teacher for access to the gymnasium can be difficult but does offer an alternative to going outdoors).

## III). Reflection

My coursework on Modern Physics, which spent time focusing on the structure of the atom, was beneficial in clearing up my misconceptions so that I could convey more accurate information to my students when designing this activity.

In the future, I would break it up over two class periods, and have the students get started on a reading assignment with a worksheet after completing part one. The second class period could be devoted to parts 2 and 3 with a more in-depth homework assignment.

This project relates very strongly to the Constructivist theory of learning because the students are actively participating in the learning process. Also, several schools of thought (such as the

Montessori philosophy) emphasize learning through play and this activity links up nicely with a play model for learning because the students were smiling, laughing, and generally seemed to be enjoying themselves.

My professional development has benefited from these activities because I now have a much more solid understanding of how effective active learning strategies can be for my students (Ueckert & Gess-Newsome, 2008). I was particularly shocked with how much better my seventh graders performed compared to my upperclassmen chemistry students.

## **Bioinformatics of the Maple Family Tree**

### **I.) Description of Activity**

This activity was designed to introduce students to the emerging field of bioinformatics. By the time the students are done with this activity they should be able to manipulate a nucleotide sequence and determine how similar it is to another nucleotide sequence, manipulate a 3-D computer model of a protein, and experience presenting their results in a formal poster presentation.

The students were first taught how to look up a gene sequence on the NCBI (National Center for Biotechnology Information) website, perform a BLAST (Basic Local Alignment Search Tool) analysis, compare sequences with Clustal W2 (multiple sequence alignment program for nucleotides or polypeptides.), and view protein crystal structures via CN3D (software for viewing and manipulating the crystal structures of biomolecules). After learning how to use the software, the students were given a mystery nucleotide sequence and they had to find out what species/gene the sequence belongs to and then compare their sequence to the same gene in similar species. Their results were compiled on a digital poster they made in Power Point and then presented to the class.

### **Materials Needed**

- Computer lab with internet connection.
- LCD projector hooked up to a computer.
- Worksheets (Appendix I)

### **II.) Results**

After implementing this activity students were able to very comfortably use the software required. As predicted, they were able to adapt to the technology and find uses for said technology that even the instructor did not know about. In particular, the students enjoyed manipulating 3-D computer models of proteins and then looking at residue characteristics (e.g.

hydrophobic vs. hydrophilic, positive vs. negative charge) since they had recently finished their unit on biochemistry. The negative outcome of this activity is that the students did not necessarily understand all of their results as they were working through the activity, particularly how to interpret the cladogram they generated. While they were able to give a rough interpretation (based on the poster presentations) most did not demonstrate the desired depth of understanding.

One section of 10<sup>th</sup> grade biology students (approximately 20 students) was affected by the activity.

### III.) Reflection

I took a class on Bioinformatics, which taught me how to use all of the software my students used in this activity.

In the future I would have example posters to show the students what I would like their final product to look like (in addition to the grading rubric). I would also spend more time front loading them with genetic theory on how proteins are synthesized, and giving the students reading assignments for homework so they could learn vocabulary outside of class (flipping the classroom) would allow for us to spend more time in lab working with the software and applying the terminology.

According to Bloom's Taxonomy, this was a difficult project for the students because it required them to work in Level 4 of the Cognitive Domain (specifically the Analysis). While operating in level 4, the students would break the project down into its various components, recognize the relationships between the parts, and reassemble them into a poster presentation (Mazo, 2013).

My professional development has benefited from these activities because designing these activities helped me to better understand some of the emerging technologies that are in the field of bioinformatics and give students a chance to work on something that is almost cutting edge. As a model of what they were doing, the students were shown a journal article that was published in 1997 where the researchers did the exact same thing I had my students do (Skepner; Krane, 1997).

This activity has the potential to be a powerful way to augment the teaching of evolution in a classroom, particularly since at the high school level evolution is primarily viewed by students to be something that only functions on the level of ecosystems and not on the level of proteins. Specifically, using bioinformatics to generate cladograms for specific proteins would allow the students to infer evolutionary relationships between species based off the just the gene sequence of their proteins. There is currently a disconnect between the use of computers in the field of biology, and the application of computers in biology education (Pezner & Shamir, 2009). While the use of bioinformatics at an undergraduate level is on the rise (Zauhar, 2001), students entering college across the country generally have not been exposed to bioinformatics (Wefer & Sheppard, 2008).

# Denaturation of Protein to Make Mozzarella Cheese

## I). Description of Activities

This activity was designed to teach 10<sup>th</sup> through 12<sup>th</sup> grade students about protein denaturation and Wisconsin history. Specifically the students were to qualitatively examine how pH influences the denaturation of protein, compare the physical properties of natured and denatured proteins, and practice the art of cheese making.

### Materials Needed:

- Lab Handout (Appendix II)
- Rennin
- Citric acid
- Milk (preferably unpasteurized and unspoiled)
- Stainless steel pot
- NaCl (Kosher pickling salt)
- Deionized water
- Stainless steel spoon to stir with
- Stainless steel knife
- Vinegar
- Baking soda

### Procedure.

1. Each lab group should make either a mixture of milk, rennin, NaCl and citric acid. This mixture was based off a standardized mozzarella cheese recipe (Carroll, 2009). Please refer to Appendix II.
2. Each lab group should decrease the pH of one mixture by adding 200 mL of vinegar, and increase the pH of another mixture with 20 g of baking soda. The third mixture should remain unaltered.
3. Have students go through the procedure to make mozzarella cheese and record the differences they observe in their final product. (By either increasing or decreasing the pH drastically, they should notice that the curds do not form).
  1. Note: Even when students do not alter the pH, they are likely to introduce errors that will cause the curds to not form. It is recommended that the instructor make their own batch of mozzarella without altering the pH so that the students are guaranteed to see what it looks like when curds form.

## II.) Results

One of the positive outcomes of this activity was that the students were able to experience in the importance of maintaining a proper pH when making cheese. The students seemed to truly be enjoying themselves as we worked through the activity. A negative outcome of this activity is that a large amount of cleanup is required and the students that did not follow the directions were disappointed because they were unable to make any curds. All of the 19 students that took Advanced Biology were affected.



### III.) Reflection

My coursework in the program helped me with preparing activities that could be easily understood and enjoyed by students. An emphasis was put on exploration in my courses at UW-RF and I transferred that same emphasis to my students, seeking input from them regarding which variables we should alter.

In the future I would simplify the experiment to increase the success rate of the students. A more straightforward approach would be to examine the effect of heat on protein denaturation instead of the effect of pH. This could be easily accomplished by using ultra-pasteurized milk (the most common kind at the grocery store) and unpasteurized milk as the two variables the students would investigate. If unpasteurized milk is unavailable, reconstituted powdered milk may be used as an alternative. Also, in the future I would give up on trying to make this experiment food safe for my students since it involved so much extra prep and cleanup. A feasible alternative would be to have the students make the cheese using regular laboratory glassware so they could still have the experience of making it, and if they wanted to eat some of it, they could eat some of the cheese that the instructor made separately using food safe methods. A benefit of this activity is that the students were exposed to a significant amount of chemistry and dairy science (Döhmers, 1995), which allows for the potential benefits of cross curricular teaching (Savage, 2011).

Based on Bloom's Taxonomy, the students that participated in this activity were able to reach level 3, Application, because they were able to successfully make cheese when they were focusing on what they were doing. None of the students were able to reach level 6, Evaluation, because no one reached the point that allowed for them to troubleshoot their mistakes while making the cheese.

My professional development has benefited by designing this activity because it has given me another way to help the students connect abstract concepts such as pH or protein folding with something that they can see, taste, and touch. Also, I learned a significant amount about cheese making while developing this activity, which I am now able to relate to other content areas while trying to help students scaffold new concepts onto old ones.

### Conclusion

All in all, the activities above represent an attempt to teach difficult concepts to students in a way that was engaging and effective. Some activities (such as the Atomic Duck Duck Goose) worked better than others, and all can be improved upon as they are used more. If there are any specific questions, or if original digital copies of the handouts (in a Word format) are desired, Robert Duerst can be contacted via email at [robert.duerst@gmail.com](mailto:robert.duerst@gmail.com).

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# Appendix 1: Handouts for Bioinformatics of Maple Family Tree Project

## Maple Family Tree Project Guidelines

### Background Information:

The field of bioinformatics involves the use of computers to analyze the large amount of information generated from the study of genetics (genomics) and the study of proteins (proteomics). Through the use of online databases and free software, many biologists are able to make comparisons in a matter of minutes, which would have taken months of work just 15 years ago.

**Objectives:** Working in small groups, combine knowledge from previous material on molecular biology with the emerging field of bioinformatics.

- Using a small amount of genetic information (between 700 and 1000 nucleotides) from a maple species to identify a gene from a specific maple species.
- Using the identified gene, construct a family tree, or cladogram, for maple species that grow in the Cornell, WI area.
- Write a 200 to 300 word report describing the importance of the gene (specifically, how does it help keep the organism alive) and the interpretation of the family tree you made.
- Compile the results into a digital poster using PowerPoint which will be presented to the class.

### Example Journal Article Similar to what you will be doing

By going to the website below you will be able to view the results of a study that is very similar to what you will be doing. The scientists in the study were interested in how species of maple trees were categorized, and to reach a conclusion they did many of the same things that you will need to do in this project.

- Title: “cpDNA of *Acer saccharum* and *Acer nigrum* are Very Similar.” Authors: Adam P. Skipner and Dan E. Krane  
([https://kb.osu.edu/dspace/bitstream/handle/1811/23754/V097N4\\_090.pdf?sequence=1](https://kb.osu.edu/dspace/bitstream/handle/1811/23754/V097N4_090.pdf?sequence=1))

## **Available Materials**

On the biology class website all of the necessary materials are available. You will need to go here to find:

- A copy of this handout
- Your group's assigned nucleotide sequence
- Grading rubric for project
- A premade list of genes from maples which you will use to construct your family tree
- A guide on how to identify your sequence using BLAST Analysis
- A guide on how to use ClustalW2 Analysis to construct your family tree
- A guide on how to use Cn3D to view the 3-D structure of proteins

## **Timeline of events:**

This project will last two weeks total, and there will be a total of 5 periods of in class time dedicated to the project

Class Period 1	Computer lab time where students are taught how to look up a sequence in the FASTA format.
Class Period 2	Computer lab time where students are taught how to perform a BLAST analysis.
Class Period 3	Computer lab time where students are taught how to perform a ClustalW2 analysis.
3-4 days to work on outside of class	
Class Period 4	Guided time in computer lab with instructor help.
2-3 days to work on outside of class	
Class Period 5	Present digital posters and discuss results as class. Short quiz on major concepts to follow

## **Suggestions:**

This is a large project and it is STRONGLY suggested that you spend at least 5 hours outside of class working on this project.

If you have questions outside of class, please ask me (Mr. Duerst) for help! I am here for you and want you to do well.

Since this is a group project, it may be helpful to self-assign different jobs to different members of the group and then combine work into a final product. (E.g. One person could really focus on the identification of the gene and researching what that gene does and why it is important while the other person focuses on making the family tree and writing an interpretation)

Even though this is a group project, do not let one member do all the work. You need to contribute!

Make sure you understand what the other group member is doing and ask them to explain their work to you. Not only will you be helping yourself, but you will be helping you group member because the process of explaining their work to you will also help them better understand what they did. Likewise, make sure you take time to explain your work/conclusions to your group member.

## Grading Rubric

	0 Points	20 Points	40 Points
<b>Family Tree</b>	Not included.	Included, but with wrong genes and/or formatting errors.	Included with correct genes and no formatting errors.
<b>Identification of nucleotide sequence source</b>	Completely wrong.	Correct gene, but wrong species.	Correct gene and species.
<b>Information in written portion</b>	Incorrect conclusions drawn from family tree, incorrect explanation of gene's importance.	Inadequate conclusion drawn from family tree or inadequate explanation of gene's importance.  (e.g. you cannot just say, "without the gene the tree would die." That may be correct in most cases, but it is inadequate. You need to state what the gene does for the organism.)	Correct interpretation drawn from family tree and adequate explanation of gene's importance.
<b>References</b>	Not included	Cited using wrong format for science class.	Cited using correct format for science class,
<b>Grammar</b>	More than 10 grammar errors on poster.	3-10 grammar errors on poster.	0-2 grammar errors on poster.
<b>Use of in Class time</b>	Student off task the majority of the in class time	Student was observed to off task during class time at least 4 times.	Student used class time effectively.

**Extra Credit:** To earn extra credit on this project, your group may decide to include a 3D image of the structure of the molecule encoded by your gene, or a similar molecule. You can search the internet and hope you get lucky, or you could use some software called Cn3D (refer to the guide titled "Manipulating Known Protein Structures using Cn3D," which is available on the class website). You may also include the all of the "Jalview" results from the ClustalW2 analysis.

	2 Points	4 Points	6 Points
<b>Molecule's 3D Structure</b>	A 3D image of a molecule was included, but it was not labeled.	A 3D image of a similar molecule was included, and it is correctly labeled.	A 3D image of a similar from the program Cn3D or an image of similar quality was included, and it is correctly labeled.
<b>JalView Results</b>	Results of ClustalW2 included with no explanation of how to interpret in the caption.	Results of ClustalW2 included, with incorrect explanation of how to interpret in the caption.	Results of ClustalW2 included with correct explanation of how to interpret in the caption.

## Genes in the *Acer* Genus

Below you will find links to specific genes from the NCBI databases that you should use to construct for family tree. For example, if your group is assigned a sequence from a gene that encodes for tRNA molecule for leucine in the chloroplast (chloroplast tRNA-Leu), you will use only that gene to construct a family tree of the different maple species and will not use any of the genes that encode for cytochrome oxidase or ribosomal RNA.

To use the links below, you will need to Ctl+click them to go to the NCBI entry for that gene. Then access the sequence in a FASTA format, and copy into a separate Word document. Repeat for the equivalent gene in each species of maple. Make sure to save this Word document you are creating!

Finally, copy all of the FASTA sequences from your Word document into the entry field for ClustalW2 and perform your analysis.

If you have questions, read the handouts titled “Using BLAST Analysis to Identify Nucleotide and Amino Acid Sequences” and “Using ClustalW Analysis to Identify Conservation in Nucleotide and Amino Acid Sequences” or ask a classmate. After trying those things, if you still need help, please ask your instructor.

### **Sugar maple**

- [Acer saccharum cytochrome oxidase subunit 1 \(cox1\) gene, partial cds; mitochondrial](#)
- [Acer saccharum 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1 and 5.8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence](#)
- [Acer saccharum voucher Li J. 6186 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit gene, partial cds; chloroplast](#)
- [Acer saccharum chloroplast tRNA-Leu gene, partial sequence; and trnL-trnF intergenic spacer region, complete sequence](#)

### **Norway Maple**

- [Acer platanoides voucher AP367 cytochrome oxidase subunit 1 \(COI\) gene, partial cds; mitochondrial](#)
- [Acer platanoides 18S ribosomal RNA gene, partial sequence, internal transcribed spacer 1, complete sequence, 5.8S ribosomal RNA gene, complete sequence, internal transcribed spacer 2, complete sequence, 26S ribosomal RNA gene, partial sequence](#)
- [Acer platanoides chloroplast partial rbcL gene for ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit, country Italy:Latium](#)

- *Acer platanoides* chloroplast tRNA-Leu gene, partial sequence; and trnL-trnF intergenic spacer region, complete sequence

### **Red Maple**

- *Acer rubrum* voucher JAG 0185 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial
- *Acer rubrum* 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5.8S ribosomal RNA gene; and internal transcribed spacer 2, complete sequence
- *Acer rubrum* ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds; chloroplast
- *Acer rubrum* chloroplast tRNA-Leu gene, partial sequence; and trnL-trnF intergenic spacer region, complete sequence

### **Box Elder**

- *Acer negundo* voucher AP244 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial
- *Acer negundo* 18S ribosomal RNA gene, partial sequence, internal transcribed spacer 1, 5.8S ribosomal RNA, and internal transcribed spacer 2 genes, complete sequences and 26S ribosomal RNA gene, partial sequence
- *Acer negundo* ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds; chloroplast
- *Acer negundo* chloroplast tRNA-Leu gene, partial sequence; and trnL-trnF intergenic spacer region, complete sequence

### **Amur Maple**

- *Acer tataricum* subsp. *ginnala* 18S ribosomal RNA gene, partial sequence, internal transcribed spacer 1, 5.8S ribosomal RNA genes, complete sequences and internal transcribed spacer 2 gene, partial sequence
- *Acer tataricum* subsp. *ginnala* ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds; chloroplast
- *Acer ginnala* chloroplast tRNA-Leu gene (partial) for transfer RNA-Leu and trnL-trnF intergenic spacer, isolate acc. 1

### **Japanese Maple**

- *Acer palmatum* subsp. *palmatum* 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5.8S ribosomal RNA gene; and internal transcribed spacer 2, complete sequence



- *Acer palmatum* ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcl) gene, partial cds; chloroplast
- *Acer palmatum* tRNA-Leu (trnL) gene and trnL-trnF intergenic spacer, partial sequence; chloroplast

## Unknown *Acer* Nucleotide Sequences

Your group will be assigned one of the below sequence numbers. The nucleotide sequence is given in a FASTA format and you will need to perform a BLAST analysis to identify the gene the sequence it came from.

If you have questions, read the handouts titled “Using BLAST Analysis to Identify Nucleotide and Amino Acid Sequences” or ask a classmate. After trying both of those things, if you still need help, please ask your instructor.

### Sequence 1

```
CTATCTGGTGTTCATCCATTTTAGGTTCTATCAATTTTATAACAACATCTCCAACATGCGTGGACCTG
GAATGACTATGCATAGATCACCCCTATTTGTGTGGTCCGTTCTAGTGACAGCATTCCCACCTTTTATTATC
ACTTCCGGTACTGGCAGGGGCAATTACCATGTTATTAACCGATCGAACTTTAATACAACCTTTTCTGAT
CCCGCTGGAGGGGAGACCCCATATTATAACCAGCATCTCTTTTGGTTCTTCGGTCATCCAGAGGTGTATA
TTCCATTCTGCCTGGATCCGGTATCATAAGTCATATCGTTTTCGACTTTTTTCGGGAAAACCGGTCTTCGG
GTATCTAGGCATGGTTTATGCCATGATCAGTATAGGTGTTCTTGGATTTCTTGTGGGCTCATCATATG
TTTACTGTGGGCTTAGACGTTGATACCCGTGCCTACTTCACCGCAGCTACCATGATCATAGCAGTCCCCA
CTGGAATCAAAATCTTTAGTTGGATCGCTACCATGTGGGGGGTTTCGATACAATACAAAACACCCATGTT
```

### Sequence 2

```
CTGCGGAAGGATCATTGTGCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTTCTATCA
TTAGGGGGGGGAGCACGGGTGCGCGAGCCTCGTGGTCCTCCTCTGCAGTCGGATCGATGGCACCCGCCGT
CGCTCCCTCGGCACAACAACGAACCCCGGCGCGGACCCGCGCAAGGAATCTTAACGACAGAGCGTGCCCC
TGCCGCCCCGAAACGGTGTGTGTGCTTGTGGCGTGCCTTCTTCCATTATTTAAAACGACTCTCGGCAA
CGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCC
CGTGAACCATCGAGTCTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGT
GTCACGCATCGTTGCCCCCCCCCTCCGAAACCCCCCTCCTCTCGAAAGAGACGATGGGACTTCGGTGTGG
GCGGATATTGGCCTCCCGTGGGCCGAACGGCTCGCGGTTGGCCTAAATCTGAGTCGTGCGCGATGGGCGC
CGTGACGTTTGGTGGTCAATAAACCTCGAGCTCTCGTGCAGCGTCCGTGTCGGTACAGTAAGGCTCAC
TGACCCTGAAGCGTTGTCAACAACGCACGCATCGCGACCCC
```

### Sequence 3

```
TGGATTCAAAGCCGGTGTAAAGATTATAAATTGACTTATTATACTCCTGACTATGTAACCAAAGATA
CTGATATCTTGGCAGCATTCCGAGTAACTCCTCAACCCGGGGTTCCGCCCAGGAAGCCGGGGCCGCGGT
AGCTGCGGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT
AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC
CTTTAGACCTTTTTGAAGAGGGTCTGTTACTAACATGTTTACTTCCATTGTGGGTAATGTATTTGGGTT
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGCGTATTGAAAACCTTTCCAAGGC
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTACTA
TTAAACCTAAATTGGGATTATCCGCTAAGAATATGGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT
TGACTTTACCAAAGATGATGAGAACGTAAACTCCAACCATTTATGCGTTGGAGAGACCGTTTCTTGTGTT
TGTGCAGAAGCTATTTTTAAATCGCAGGCAGAACTGGTGAATCAAAGGTCATTACTTGAATGCTACTG
CAGGTACATGGGAAGAGATGCTAAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCCATCGTAATGCA
TGACTACTTAAACAGGGGATTACCCGCAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT
CTTCACATCCATCGTGCAATGCATGCAGTTATTGATAGACAGAAGTATCATGGTATACACTTTCGTGTAC
```

TAGCTAAAGCTTTGCGTATGTCTGGTGGAGATCATATTCACGCAGGTACAGTAGTAGGTAAACTTGAAGG  
AGAAAGAGACATAAAGCTTTGGGCTTTGTTGATTTACTACGTGATGATTTTTATTGAAAAAGACCGAAGCCGC  
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCCTGGCTTCAGGGGGTATTACAG  
TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGTAAGTCAATTCGGTGGAGGAACCTTT  
AGGACACCCTTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT  
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATTTCGTGAGGCTAGCAAATGGAGTGCTG  
AATTGGCTGCTGCTTGTGAAGTATGG

#### Sequence 4

TTTCAAATTCAGAGAAACCCGGGAATCAAAAATGGGCAATCCTG  
AGCCAAATCCTGTTTTACGAGAATAAAACAAAGCAAACAAGGGTTTCAGAAAGCGAGAAAAGGGATAGGTG  
CAGAGACTCAACGGAAGCTGTTCTAACAAATGGAGTTGACTGCCTTTTTTTGGGAAAGAAAGGAAAATTA  
ATCGAATATCGAAACTGCATAAAGGATGAAGGATAAGTCTATAATACACTATGGATACAAAATGAAAAAT  
TATCTCAAAAATGACAACCTGAAGCTGTATTTATATTAATGAAAAAGAAAACAAAAGAATTGGGATTCCA  
AGTTGAAGAATCGAATATTCATTGATCAAATCATTCACTCCATAGTCTGGTAGATCGTTTTCTTTTGAAGA  
ACTGATTACTCAGACGAGAATAAAGATAGAGTCCCATTCTACATGTCAATATAAATACCGGCAACAATGA  
AATTTATAGTAAGAGGAAAATCCGTCGACTTTAGAAATCGTGAGGGTTCAAGTCCCTCTATCCCCCAA  
AGGACCGAATTTGACTATTTCTCCTACCCTCTCTTTTTTTTAGTGGTTCAAATTCGTTCTCTTTCTCATT  
CATTCTACCTTTTTTACAAACGTATCAGAGCAGAATTTTTTTTTCTTTTTATCACAATCACAAGTGGTGTGGT  
ATATATGATATACGTACAAATGAACATCTTTGAACAAAGAATCCCGATTACAATTCATATCGTTGCTCA  
TACTGAAACTTATAAATTATTCTATTCAAGAAATCGAATTCACGCTCAAGACTTTTAATACTTTTTTTA  
TCTTTTTTAATTGACATAGACCCCCAGTCATCTAGTAAAATGAGGATGATGCGCCGGTAATGGTCTGGGATA  
GCTCAGCTGGTAGAGCAGAGGAC

#### Sequence 5

CATTGTGCAAACCTGCCAGCAGAACGACCCGCGAAGTTCGTTTTATCACCAGGGGAAGCAC  
GGGTGCGCGAGCCTCGTGTTCCCCCTTTCAGTTGGATCGTTGGCACCTGCCCTCGCTCCCTCGGCACAA  
CAACGAACCCCGGCGCGGACCGCGCAAGGAAAATAACAGGGGAGCGTGCCCTTGCCGCCCCGAAACG  
GTGTGCGTGCCTGTGGCATTGCCTTCTTTTCAATTTATTTAAACGACTCTCGGCAACGGATATCTCGGCTC  
TCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGTGAACCATCGAGT  
CTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGTGTACGCATCGTTGC  
CCCCCAAACCCCTTTTACCCCACTTTTC

#### Sequence 6

GTAAAGATTATAAATTGACTTATTATACTCCTGAGTATGTAACCAAAGATA  
CTGATATCTTGGCAGCATTCCGAGTAACTCCTCAACCCGGGGTTCCGCCCCGAGGAAGCCGGGGCCGCGGT  
AGCTGCGGAATCTTCTACTGGTACATGGACAACGTGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT  
AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC  
CTTTAGACCTTTTTGAAGAGGGTCTGTTACTAACATGTTTACTTCCATTGTGGGTAATGTATTTGGGTT  
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGCTATTTCGAAAACCTTTCAAGGC  
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTA  
TTAAACCTAAATTGGGATTATCCGCTAAGAACTACGGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT  
TGACTTTACCAAAGATGATGAGAACGTAACCTCCAACATTTATGCGTTGGAGAGACCGTTTTCTGTTTT  
TGTGCAGAAGCTATTTATAAATCGCAGGCAGAACTGGTGAAATCAAAGGTCATTACTTGAATGCTACTG  
CAGGTACATGGGAAGAGATGCTAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCCATCGTAATGCA  
TGACTACTTAAACAGGGGATTACCCGCAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT  
CTTCACATCCATCGTGCAATGCATGCAGTTATTGATAGACAGAAGAATCATGGTATACACTTTTCGTGTAC  
TAGCTAAAGCTTTGCGTATGTGAGGTGGAGATCATATTCACGCAGGTACAGTAGTAGGTAAACTTGAAGG  
AGAAAGAGACATAAAGCTTTGGGCTTTGTTGATTTACTACGTGATGATTTTTATTGAAAAAGACCGAAGCCGC  
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCCTGGCTTCAGGGGGTATTACAG

TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGTACTGCAATTCGGTGGAGGAACTTT  
AGGACACCCTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT  
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATCCGTGAGGCTAGCAAATGGAGTGCTG  
AATTGGCTGCTGCTTGTGAAATATGGAAGGAGATCAAATTTGAATTTGAAGCAATGGATACTTTGTAATC  
CAGTGATTAACGTTTGGTTTATTAGTTGAATTG

### **Sequence 7**

GTAGGTGAACCTGCGGAAGGATCATTGTGCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTTCTATCA  
TTAGGGGGGGGAGCACGGGTGCGCGAGCCTCGTGGTCTCTCTGCAGTCGGATCGATGGCACCCGCCGT  
CGCTCCCTCGGCACAACAACGAACCCCGGCGCGGACCGCGCAAGGAATCTTAACGACAGAGCGTGCCCC  
TGCCGCCCGGAAACGGTGTGTGTGCTTGTGGCGTGCCTTCTTCCATTATTTAAAACGACTCTCGGCAA  
CGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCC  
CGTGAACCATCGAGTCTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGT  
GTCACGCATCGTTGCCCCCCCCCTCCGAAACCCCCCTCTCTCGAAAGAGACGATGGGACTTCGGTGTGG  
GCGGATATTGGCCTCCCGTGGGCCGAACGGCTCGCGGTTGGCCTAAATCTGAGTCGTGGCGATGGGCGC  
CGTGACGTTCCGTGGTCAATAAACCTCGAGCTCTCGTCGCGCGTCCGTCTCGGTACAGTAAGGCTCAC  
TGACCCTGAAGCGTTGTCAACAACGC

### **Sequence 8**

GTGTTTTATCCATTTTAGGTTCTATCAATTTTATAACAACATCTCCAACATGCGTGGACCTG  
GAATGACTATGCATAGATCACCCCTATTTGTGTGGTCCGTTCTAGTGACAGCATTCCCCTTTTATTATC  
ACTTCCGGTACTGGCAGGGGCAATTACCATGTTATTAACCGATCGAACTTTAATACAACCTTTTCTGAT  
CCCGCTGGAGGGGAGACCCCATATTATAACCAGCATCTCTTTTGGTTCTTCGGTCATCCAGAGGTGTATA  
TTCCCATTTCTGCCTGGATCCGGTATCATAAGTCATATCGTTTTCGACTTTTTTCGGGAAAACCGGTCTTCGG  
GTATCTAGGCATGGTTTTATGCCATGATCAGTATAGGTGTTCTTGGATTTCTTGTGGGCTCATCATATG  
TTTACTGTGGGCTTAGACGTTGATACCCGTGCCTACTTACCAGCAGCTACCATGATCATAGCAGTCCCCA  
CTGGAATCAAATCTTTAGTTGGATCGCTACCATGTGGGGGGTTTCGATACAATACAAAACACCCATGTT  
ATTTGCTGT

### **Sequence 9**

CAGCATTCCGAGTAACTCCTCAACCCGGGGTCCGCCCGAGGAAGCCGGGGCCGCGGT  
AGCTGCGGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT  
AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC  
CTTTAGACCTTTTTGAAGAGGGTCTGTACTAATCATGTTTACTTCCATTGTGGGTAATGTATTTGGGT  
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGCGTATTTCGAAAACCTTTCCAAGG  
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTACTA  
TTAAACCTAAATTTGGGATTATCCGCTAAGAACATGGTGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT  
TGACTTTACCAAAGATGATGAGAACGTAAACTCCCAACATTTATGCGTTGGAGAGACCGTTTCTTGT  
TGTGCAGAAGCTATTTTTAAATCGCAGGCAGAACTGGTGAATCAAAGGTCATTACTTGAATGCTACTG  
CAGGTACATGGGAAGAGATGCTAAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCTATCGTAATGCA  
TGACTACTTAAACAGGGGATTACCCGCAAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT  
CTTACATCCATCGTGCAATGCATGCAGTTATTGATAGACAGAAGAATCATGGTATACACTTTTCGTGTAC  
TAGCTAAAGCTTTGCGTATGTCTGGTGGAGATCATATTCACGCAGGTACAGTAGTAGGTAAACTTGAAGG  
AGAAAGAGACATAACTTTGGGCTTTGTTGATTTACTACGTGATGATTTTATTGAAAAAGACCGAAGCCGC  
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCGTGGCTTCAGGGGGTATTACAG  
TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGTACTGCAATTCGGTGGAGGAACTTT  
AGGACACCCTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT  
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATTCGTGAGGCTAGCAAATGGAGTGCTG  
AATTGGCTGCTGCTTGTGAAGTATGGAAGGAGATCAAATTTGAATTTGAAGCAATGGATACTTTGTAATC  
CAGTGATTAACGTTTGGTTTATTAGTTGAATTG

**Sequence 10**

TATGGAAACCTACTAAGTGATAACTTTCAAATTCAGAGAAACCCGGAATCAAAAATGGGCAATCCTG  
AGCCAAATCCTGTTTTACGAGAATAAAACAAAGCAAACAAGGGTTCAGAAAGCGAGAAAAGGGATAGGTG  
CAGAGACTCAACGGAAGCTGTTCTAACAAATGGAGTTGACTGCCTTTTTTTGGGAAAGAAAGGAAAATTA  
ATCGAATATCGAAACTGCATAAAGGATGAAGGATAAGTCTATAATACACTATGGATACAAAATGAAAAAT  
TATCTCAAAAATGACAACCTGAAGCTGTATTTATATTAATGAAAAAGAAAACAAAAGAATTGGGATTCCA  
AGTTGAAGAATCGAATATTCATTGATCAAATCATTCACTCCATAGTCTGGTAGATCGTTTTCTTTGAAGA  
ACTGATTACTCAGACGAGAATAAAGATAGAGTCCCATTCTACATGTCAATATAAATACCGCAACAATGA  
AATTTATAGTAAGAGGAAAATCCGTCGACTTTAGAAATCGTGAGGGTTCAGTCCCTCTATCCCCCAA  
AGGACCGATTTGACTATTTCTCCTACCCTCTCTTTTTTTTTAGTGGTTCAAATTCGTTCTCTTTCTCATT  
CATTCTACTTTTTACAAACGTATCAGAGCAGAATTTTTTTTTCTTTTATCACAATCACAAGTGGTGTGGT  
ATATATGATATACGTACAAATGAACATCTTTGAACAAAGAATCCCGATTACAAATTCATATCGTTGCTCA  
TACTGAACTTTATAAATTATTCTATTCAAGAAATCGAATTCACGCTCAAGACTTTTAATACTTTTTTTA  
TCTTTTTTAATTGA

**Sequence 11**

GAAGGATCATTGTGCAACCTGCCAGCAGAACGACCCGCGAACTCGTTTTATCACCAGGGGAAGCAC  
GGGTGCGCGAGCCTCGTGTTCCTCCCTTTGCAGTTGGATCGTTGGCACCTGCCCTCGCTCCCTCGGCACAA  
CAACGAACCCCGCGCGGACCGCGCAAGGAAAATAACAGGGGAGCGTGCCTTGCCTCCCGGAAACG  
GTGTGCGTGCCTGTGGCATTGCCTTCTTTTATTTAAAACGACTCTCGGCAACGGATATCTCGGCTC  
TCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGTGAACCATCGAGT  
CTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGTGTCACGCATCGTTGC  
CCCCCAAACCCCTTTTACCCC

**Sequence 12**

GGATTCAAAGCCGGTGTAAAGATTATAAATTGACTTATTATACTCCTGAGTATGTAACCAAAGATA  
CTGATATCTTGGCAGCATTCCGAGTAACTCCTCAACCCGGGGTTCGCCCCGAGGAAGCCGGGGCCGCGGT  
AGCTGCGGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT  
AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC  
CTTTAGACCTTTTTGAAGAGGGTCTGTTACTAACATGTTTACTTCCATTGTGGGTAATGTATTTGGGTT  
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGCGTATTTCGAAAACCTTTCCAAGGC  
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTACTA  
TTAAACCTAAATTGGGATTATCCGCTAAGAACTACGGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT  
TGACTTTACCAAAGATGATGAGAACGTAAACTCCCAACCTTTATGCGTTGGAGAGACCGTTTCTTGTTT  
TGTGCAGAAGCTATTTATAAATCGCAGGCAGAACTGGTGAATCAAAGGTCACTTGAATGCTACTG  
CAGGTACATGGGAAGAGATGCTAAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCCTATCGTAATGCA  
TGACTACTTAACAGGGGATTACCCGCAAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT  
CTTCACATCCATCGTGCAATGCATGCAGTTATTGATAGACAGAAGAATCATGGTATACACTTTTCGTGTAC  
TAGCTAAAGCTTTGCGTATGTGAGGTGGAGATCATATTCACGCAGGTACAGTAGTAGGTAAACTTGAAGG  
AGAAAGAGACATAACTTTGGGCTTTGTTGATTTACTACGTGATGATTTTATTGAAAAAGACCGAAGCCGC  
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCGTGGCTTCAGGGGGTATTACAG  
TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGTACTGCAATTCGGTGGAGGAACCTT  
AGGACACCCTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT  
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATCCGTGAGGCTAGCAAATGGAGTGCTG  
AATTGGCTGCTGCTTGTGAAATATGGAAGGAGATCAAATTTGAATTTGAAGCAATGGATACTTTGTAATC  
CAGTGATTAACGTTTGGTTTATT ]

## Unknown *Acer* Nucleotide Sequences (ANSWER KEY)

Your group will be assigned one of the below sequence numbers. The nucleotide sequence is given in a FASTA format and you will need to perform a BLAST analysis to identify the gene the sequence it came from.

If you have questions, read the handout titled "Using BLAST Analysis to Identify Nucleotide and Amino Acid Sequences" or ask a classmate. After trying both of those things, if you still need help, please ask your instructor.

### Sequence 1

[\(\*Acer saccharum\* cytochrome oxidase subunit 1 \(cox1\) gene, partial cds; mitochondrial\)](#)

```
CTATCTGGTGTTCATCCATTTTAGGTTCTATCAATTTTATAACAACATCTCCAACATGCGTGGACCTG
GAATGACTATGCATAGATCACCCCTATTTGTGTGGTCCGTTCTAGTGACAGCATTCCCACCTTTTATTATC
ACTTCCGGTACTGGCAGGGGCAATTACCATGTTATTAACCGATCGAACTTTAATACAACCTTTTCTGAT
CCCGCTGGAGGGGGAGACCCCATATTATAACAGCATCTCTTTTGGTTCTTCGGTCATCCAGAGGTGTATA
TTCCCATTTCTGCCTGGATCCGGTATCATAAGTCATATCGTTTTCGACTTTTTTCGGGAAAACCGGTCTTCGG
GTATCTAGGCATGGTTTATGCCATGATCAGTATAGGTGTTCTTGGATTTCTTGTTTGGGCTCATCATATG
TTTACTGTGGGCTTAGACGTTGATACCCGTGCCTACTTCACCGCAGCTACCATGATCATAGCAGTCCCCA
CTGGAATCAAATCTTTAGTTGGATCGCTACCATGTGGGGGGTTTCGATACAATACAAAACACCCATGTT
```

### Sequence 2

[\(\*Acer platanoides\* 18S ribosomal RNA gene, partial sequence, internal transcribed spacer 1, complete sequence, 5.8S ribosomal RNA gene, complete sequence, internal transcribed spacer 2, complete sequence, 26S ribosomal RNA gene, partial sequence\)](#)

```
CTGCGGAAGGATCATTGTGCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTTCTATCA
TTAGGGGGGGGAGCACGGGTGCGCGAGCCTCGTGGTCTCCTCTGCAGTCGGATCGATGGCACCCGCCGT
CGCTCCCTCGGCACAACAACGAACCCCGGCGCGGACCGCGCCAAGGAATCTTAACGACAGAGCGTGCCCC
TGCCGCCCCGAAACGGTGTGTGTGCTTGTGGCGTCGCCTTCTTCCATTATTTAAAACGACTCTCGGCAA
CGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCC
CGTGAACCATCGAGTCTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGT
GTCACGCATCGTTGCCCCCCCCCTCCGAAACCCCTCCTCTCGAAAGAGACGATGGGACTTCGGTGTGG
GCGGATATTGGCCTCCCGTGGGCGCAACGGCTCGCGGTTGGCCTAAATCTGAGTCGTGGCGATGGGCGC
CGTGACGTTTCGGTGGTCAATAAACCTCGAGCTCTCGTTCGCGGTCCGTCGTGGTACAGTAAGGCTCAC
TGACCCTGAAGCGTTGTCAACAACGCACGCATCGCGACCC
```

### Sequence 3

[\(\*Acer rubrum\* ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit \(rbcL\) gene, partial cds; chloroplast\)](#)

```
TGGATTCAAAGCCGGTGTAAAGATTATAAATTGACTTATTATACTCCTGACTATGTAACCAAAGATA
CTGATATCTTGGCAGCATTCCGAGTAACTCCTCAACCCGGGGTTCCGCCCCGAGGAAGCCGGGGCCGCGGT
AGCTGCGGAATCTTCTACTGGTACATGGACAACGTGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT
```

AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC  
CTTTAGACCTTTTTGAAGAGGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTAATGTATTTGGGTT  
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGCTATTTCGAAAACCTTTCCAAGGC  
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTACTA  
TTAAACCTAAATTGGGATTATCCGCTAAGAAGTATGGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT  
TGACTTTACCAAAGATGATGAGAACGTAACCTCCAACCTTTATGCGTTGGAGAGACCGTTTCTTGTTT  
TGTGCAGAAGCTATTTTTAAATCGCAGGCAGAACTGGTGAATCAAAGGTCATTACTTGAATGCTACTG  
CAGGTACATGGGAAGAGATGCTAAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCCTATCGTAATGCA  
TGACTACTTAACAGGGGGATTACCCGCAAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT  
CTTCACATCCATCGTGCATGCATGCAGTTATTGATAGACAGAAGAATCATGGTATACACTTTCTGTGTAC  
TAGCTAAAGCTTTGCGTATGTCTGGTGGAGATCATATTCACGCAGGTACAGTAGGTAAACTTTGAAGG  
AGAAAGAGACATAACTTTGGGCTTTGTTGATTTACTACGTGATGATTTTTATTGAAAAGACCGAAGCCGC  
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCGTGGCTTCAGGGGGTATTACAG  
TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGTAAGTGCATTCGGTGGAGGAACCTT  
AGGACACCCTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT  
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATTTCGTGAGGCTAGCAAATGGAGTGCTG  
AATTGGCTGCTGCTTGTGAAGTATGG

#### **Sequence 4**

([Acer negundo chloroplast tRNA-Leu gene, partial sequence; and trnL-trnF intergenic spacer region, complete sequence](#))

TTTCAAATTCAGAGAAACCCGGGAATCAAAAATGGGCAATCCTG  
AGCCAAATCCTGTTTTACGAGAATAAAACAAAGCAAACAAGGGTTTCAGAAAGCGAGAAAAGGGATAGGTG  
CAGAGACTCAACGGAAGCTGTTCTAACAATGGAGTTGACTGCCTTTTTTTGGGAAAGAAAGGAAAATTA  
ATCGAATATCGAAAAGTGCATAAAGGATGAAGGATAAGTCTATAATACACTATGGATACAAAATGAAAAAT  
TATCTCAAAAATGACAACCTGAAGCTGTATTTATATTAATGAAAAGAAAACAAAAGAATTGGGATTCCA  
AGTTGAAGAATCGAATATTCATTGATCAAATCATTCACTCCATAGTCTGGTAGATCGTTTTCTTTTGAAGA  
ACTGATTACTCAGACGAGAATAAAGATAGAGTCCCATTCTACATGTCAATATAAATACCGGCAACAATGA  
AATTTATAGTAAGAGGAAAATCCGTCGACTTTAGAAATCGTGAGGGTTCAAAGTCCCTCTATCCCCCAA  
AGGACCGATTTGACTATTTCTCCTACCCTCTCTTTTTTTTTAGTGGTTCAAATTCGTTCTCTTTCTCATT  
CATTCTACCTTTTTTACAAACGTATCAGAGCAGAATTTTTTTTTCTTTTATCACAATCACAAGTGGTGTGGT  
ATATATGATATACGTACAAATGAACATCTTTGAACAAAGAATCCCGATTACAAATTCATATCGTTGCTCA  
TACTGAAACTTATAAATTATTCTATTCAAGAAATCGAATTCACGCTCAAGACTTTTAATACTTTTTTTA  
TCTTTTTTAATTGACATAGACCCAGTCATCTAGTAAAATGAGGATGATGCGCCGGTAATGGTCGGGATA  
GCTCAGCTGGTAGAGCAGAGGAC

#### **Sequence 5**

([Acer tataricum subsp. ginnala 18S ribosomal RNA gene, partial sequence, internal transcribed spacer 1, 5.8S ribosomal RNA genes, complete sequences and internal transcribed spacer 2 gene, partial sequence](#))

CATTGTGCAAACCTGCCAGCAGAACGACCCGCGAACTCGTTTTATCACCAGGGGAAGCAC  
GGGTGCGCGAGCCTCGTGTTCCCTTTGTCAGTTGGATCGTTGGCACCTGCCCTCGCTCCCTCGGCACAA  
CAACGAACCCCGGCGCGGACCGCGCAAGGAAAATAACAGGGGAGCGTGCCCTTGCCGCCCCGAAACG  
GTGTGCGTGCCTGTGGCATTGCCTTCTTTTCAATTTATTTAAACGACTCTCGGCAACGGATATCTCGGCTC  
TCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGTGAACCATCGAGT  
CTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGTGTACGCATCGTTGC  
CCCCCAAACCCCTTTTACCCCACTTTTC

### Sequence 6

(*Acer palmatum* ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds; chloroplast)

```
GTAAAGATTATAAATTGACTTATTATACTCCTGAGTATGTAACCAAAGATA
CTGATATCTTGGCAGCATTCCGAGTAACTCCTCAACCCGGGGTTCGCCCCGAGGAAGCCGGGGCCGCGGT
AGCTGCGGAATCTTCTACTGGTACATGGACAACGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT
AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC
CTTTAGACCTTTTTGAAGAGGGTCTGTTACTAACATGTTTACTTCCATTGTGGGTAATGTATTTGGGTT
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGGTATTGAAAACCTTCCAAGGC
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTACTA
TTAAACCTAAATTGGGATTATCCGCTAAGAACTACGGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT
TGACTTTACCAAAGATGATGAGAACGTAAACTCCCAACATTTATGCGTTGGAGAGACCGTTTCTTGTTT
TGTGCAGAAGCTATTTATAAATCGCAGGCAGAACTGGTGAAATCAAAGGTCATTACTTGAATGCTACTG
CAGGTACATGGGAAGAGATGCTAAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCCTATCGTAATGCA
TGACTACTTAAACAGGGGATTACCCGCAAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT
CTTCACATCCATCGTGCAATGCATGCAGTTATTGATAGACAGAAGAATCATGGTATACACTTTTCGTGTAC
TAGCTAAAGCTTTGCGTATGTCAGGTGGAGATCATATTCACGCAGGTACAGTAGTAGGTAACCTGAAGG
AGAAAGAGACATAAATTTGGGCTTTGTTGATTTACTACGTGATGATTTTATTGAAAAGACCCGAAGCCGC
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCGTGGCTTCAGGGGGTATTACAG
TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGTACTGCAATTCGGTGGAGGAACCTT
AGGACACCCTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATCCGTGAGGCTAGCAAATGGAGTGCTG
AATTGGCTGCTGCTTGTGAAATATGGAAGGAGATCAAATTTGAATTTGAAGCAATGGATACTTTGTAATC
CAGTGATTAACGTTTGGTTTATTAGTTGAATTG
```

### Sequence 7

(*Acer platanoides* 18S ribosomal RNA gene, partial sequence, internal transcribed spacer 1, complete sequence, 5.8S ribosomal RNA gene, complete sequence, internal transcribed spacer 2, complete sequence, 26S ribosomal RNA gene, partial sequence)

```
GTAGGTGAACCTGCGGAAGGATCATTGTGCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTTCTATCA
TTAGGGGGGGGAGCACGGGTGCGCGAGCCTCGTGGTCCCTCCTCTGCAGTCGGATCGATGGCACCCGCCGT
CGCTCCCTCGGCACAACAACGAACCCCGGCGCGGACCGCGCAAGGAATCTTAACGACAGAGCGTGCCCC
TGCCGCCCGGAAACGGTGTGTGTGCTTGTGGCGTGCCTTCTTCCATTATTTAAAACGACTCTCGGCAA
CGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCC
CGTGAACCATCGAGTCTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGT
GTCACGCATCGTTGCCCCCCCCCTCCGAAACCCCCCTCCTCTCGAAAGAGACGATGGGACTTCGGTGTGG
GCGGATATTGGCCTCCCGTGGGCGCAACGGCTCGCGGTTGGCCTAAATCTGAGTCGTGCGGATGGGCGC
CGTGACGTTCCGGTGGTCAATAAACCTCGAGCTCTCGTTCGCGCGTCCGTGCTCGGTACAGTAAGGCTCAC
TGACCCTGAAGCGTTGTCAACAACGC
```

### Sequence 8

(*Acer saccharum* cytochrome oxidase subunit 1 (cox1) gene, partial cds; mitochondrial)

```
GTGTTTCATCCATTTTAGGTTCTATCAATTTTATAACAACATCTCCAACATGCGTGGACCTG
GAATGACTATGCATAGATCACCCCTATTTGTGTGGTCCGTTCTAGTGACAGCATTCCCACTTTTATTATC
ACTTCCGGTACTGGCAGGGGCAATTACCATGTTATTAACCGATCGAAACTTTAATACAACCTTTTCTGAT
```



CCCGCTGGAGGGGGAGACCCCATATTATAACCAGCATCTCTTTTGGTTCTTCGGTCATCCAGAGGTGTATA  
TTCCCATTTCTGCCTGGATCCGGTATCATAAGTCATATCGTTTTCGACTTTTTCGGGAAAACCGGTCTTCGG  
GTATCTAGGCATGGTTTTATGCCATGATCAGTATAGGTGTTCTTGGATTTCTTGTGGGCTCATCATATG  
TTTACTGTGGGCTTAGACGTTGATACCCGTGCCTACTTCACCGCAGCTACCATGATCATAGCAGTCCCCA  
CTGGAATCAAAATCTTTAGTTGGATCGCTACCATGTGGGGGGTTTCGATACAATACAAAACACCCATGTT  
ATTTGCTGT

### Sequence 9

[\(Acer rubrum ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit \(rbcl\) gene, partial cds; chloroplast\)](#)

CAGCATTCCGAGTAACTCCTCAACCCGGGGTCCGCCCGAGGAAGCCGGGGCCGCGGT  
AGCTGCGGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT  
AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC  
CTTTAGACCTTTTTGAAGAGGGTCTGTTACTAACATGTTTACTTCCATTGTGGGTAATGTATTTGGGTT  
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGCTATTGAAAACCTTTCCAAGGC  
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTACTA  
TTAAACCTAAATTGGGATTATCCGCTAAGAACTATGGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT  
TGACTTTACCAAAGATGATGAGAACGTAACCTCCAACCTTTATGCGTTGGAGAGACCGTTTCTTGT  
TGTGCAGAAGCTATTTTTAAATCGCAGGCAGAACTGGTGAATCAAAGGTCATTACTTGAATGCTACTG  
CAGGTACATGGGAAGAGATGCTAAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCCTATCGTAATGCA  
TGACTACTTAACAGGGGATTACCCGCAAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT  
CTTCACATCCATCGTGCAATGCATGCAGTTATTGATAGACAGAAGAATCATGGTATACACTTTTCGTGTAC  
TAGCTAAAGCTTTGCGTATGTCTGGTGGAGATCATATTCACGCAGGTACAGTAGTAGGTAACTTGAAGG  
AGAAAGAGACATAAATTTGGGCTTTGTTGATTTACTACGTGATGATTTTATTGAAAAAGACCGAAGCCGC  
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCGTGGCTTCAGGGGGTATTACG  
TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGCTACTGCAATTCGGTGGAGGAACCTT  
AGGACACCCTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT  
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATTTCGTGAGGCTAGCAAATGGAGTGCTG  
AATTGGCTGCTGCTTGTGAAGTATGGAAGGAGATCAAATTTGAATTTGAAGCAATGGATACTTTGTAATC  
CAGTGATTAACGTTTGGTTTATTAGTTGAATTG

### Sequence 10

[\(Acer negundo chloroplast tRNA-Leu gene, partial sequence; and trnL-trnF intergenic spacer region, complete sequence\)](#)

TATGGAAACCTACTAAGTGATAACTTTCAAATTCAGAGAAACCCGGGAATCAAAAATGGGCAATCCTG  
AGCCAAATCCTGTTTTACGAGAATAAAAACAAAGCAAACAAGGGTTTCAGAAAGCGAGAAAAGGGATAGGTG  
CAGAGACTCAACGGAAGCTGTTCTAACAAATGGAGTTGACTGCCTTTTTTTTTGGGAAAAGAAAGAAAATTA  
ATCGAATATCGAAACTGCATAAAGGATGAAGGATAAGTCTATAATACACTATGGATACAAAATGAAAAAT  
TATCTCAAAAATGACAACCTGAAGCTGTATTTATATTAATGAAAAAGAAAACAAAAGAATTGGGATTCCA  
AGTTGAAGAATCGAATATTCATTGATCAAATCATTCACTCCATAGTCTGGTAGATCGTTTCTTTTGAAGA  
ACTGATTACTCAGACGAGAATAAAGATAGAGTCCCATTCTACATGTCAATATAAATACCGGCAACAATGA  
AATTTATAGTAAGAGGAAAATCCGTCGACTTTAGAAATCGTGAGGGTTCAAGTCCCTCTATCCCCC  
AGGACCGATTTGACTATTTCTCCTACCCTCTCTTTTTTTTTTAGTGGTTCAAATTCGTTCTCTTTTCAAT  
CATTCTACCTTTTTTACAAACGTATCAGAGCAGAATTTTTTTTTCTTTTATCACAATCACAAGTGGTGTGGT  
ATATATGATATACGTACAAATGAACATCTTTGAACAAAGAATCCCGATTACAATTCATATCGTTGCTCA  
TACTGAAACTTATAAATTATTCTATTCAAGAAATCGAATTCACGCTCAAGACTTTTAAATACTTTTTTTA  
TCTTTTTTAATTGA

### Sequence 11

[\(Acer tataricum subsp. ginnala 18S ribosomal RNA gene, partial sequence, internal transcribed spacer 1, 5.8S ribosomal RNA\)](#)

genes, complete sequences and internal transcribed spacer 2  
gene, partial sequence)

GAAGGATCATTGTGCGAAACCTGCCAGCAGAACGACCCGCGAACTCGTTTTATCACCAGGGGAAGCAC  
GGGTGCGCGAGCCTCGTGTTCCTCCCTTTGCAGTTGGATCGTTGGCACCTGCCCTCGCTCCCTCGGCACAA  
CAACGAACCCCGGCGCGGACCGCGCCAAGGAAAATAACAGGGGAGCGTGCCCTTGCCGCCCCGAAACG  
GTGTGCGTGCCTGTGGCATTGCCTTCTTTCAATTTATTTAAAACGACTCTCGGCAACGGATATCTCGGCTC  
TCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCCTGAACCATCGAGT  
CTTTGAACGCAAGTTGCGCCCCAAGCCGTTAGGCCGAGGGCACGCCTGCCTGGGTGTCACGCATCGTTGC  
CCCCCAAACCCCTTTTACCCC

**Sequence 12**

(Acer palmatum ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit  
(rbcL) gene, partial cds; chloroplast)

GGATTCAAAGCCGGTGTAAAGATTATAAATTGACTTATTATACTCCTGAGTATGTAACCAAAGATA  
CTGATATCTTGGCAGCATTCCGAGTAACTCCTCAACCCGGGGTTCGCCCCGAGGAAGCCGGGGCCGCGGT  
AGCTGCGGAATCTTCTACTGGTACATGGACAACGTGTGTGGACCGATGGGCTTACCAGCCTTGATCGTTAT  
AAAGGACGATGCTACAACATTGAGCCCGTTGCTGGAGAAGAAAATCAATATATATGTTATGTAGCTTACC  
CTTTAGACCTTTTTGAAGAGGGTCTGTTACTAACATGTTTACTTCCATTGTGGGTAATGTATTTGGGTT  
TAAAGCCCTGCGCGCTCTACGTCTAGAGGATCTACGAATCCCTCCCGCGTATTGAAAACCTTTCCAAGGC  
CCCCCTCATGGCATCCAAGTTGAGAGAGATAAATTGAACAAGTATGGGCGCCCCCTATTGGGATGTACTA  
TTAAACCTAAATTGGGATTATCCGCTAAGAACTACGGTAGAGCAGTTTATGAATGTCTACGTGGTGGACT  
TGACTTTACCAAAGATGATGAGAACGTAAACTCCAACCATTTATGCGTTGGAGAGACCGTTTCTTGTTT  
TGTGCAGAAGCTATTTATAAATCGCAGGCAGAACTGGTGAAATCAAAGGTCATTACTTGAATGCTACTG  
CAGGTACATGGGAAGAGATGCTAAAAGGGCGGTATTTGCCAGAGAGTTGGGAGTTCCTATCGTAATGCA  
TGACTACTTAACAGGGGGATTACCGCAAATACTAGCTTGGCTCATTATTGCCGAGATAATGGTCTACTT  
CTTCACATCCATCGTGCAATGCATGCAGTTATTGATAGACAGAAGAATCATGGTATACACTTTTCGTGTAC  
TAGCTAAAGCTTTGCGTATGTCAGGTGGAGATCATATTCACGCAGGTACAGTAGTAGGTAACCTTGAAGG  
AGAAAGAGACATAACTTTGGGCTTTGTTGATTTACTACGTGATGATTTTATTGAAAAAGACCGAAGCCGC  
GGTATTTATTTCACTCAAGATTGGGTCTCTTTACCAGGTGTTCTGCCCGTGGCTTCAGGGGGTATTACG  
TTTGGCATATGCCTGCTTTGACCGAGATCTTTGGGGACGATTCCGTAATGCAATTCCGTGGAGGAACTTT  
AGGACACCCTTGGGGAAATGCGCCAGGCGCCGTAGCTAATCGAGTAGCTCTAGAAGCATGTGTACAAGCT  
CGTAATGAAGGACGCGATCTTGCTCGCGAGGGTAATGAAATTATCCGTGAGGCTAGCAAATGGAGTGCTG  
AATTGGCTGCTGCTTGTGAAATATGGAAGGAGATCAAATTTGAATTTGAAGCAATGGATACTTTGTAATC  
CAGTGATTAACGTTTGGTTTATT ]

## Appendix 2: Handouts Denaturation of Protein Activity

### Cheese Making Lab

Background: When making cheese, the target protein is called Casein. The reason this protein is targeted is because it makes up the majority of the protein content in milk. To cause this protein to denature, mild heat is applied while the pH is lowered slightly. This causes the hydrophobicity of several of the functional R-Groups of the amino acids to be reversed, and the structure of Casein is altered. To further aide in the Casein denaturation, a cocktail of enzymes called rennet is added. This mixture of enzymes was originally isolated from the stomach of a mammal, and helps to break down the proteins and fat present in milk.

During this lab you will alter the pH of the recipe by adding vinegar or baking soda to examine the effect that pH has on the denaturation of the casein protein.

Safety: No special concerns. Use common sense while around a stovetop burner.

Note: The instructions below were modified from a recipe found at [www.cheesemaking.com](http://www.cheesemaking.com)

Total Time: 30 minutes

Yields: Approximately ½ pound of cheese

#### Ingredients:

- 1 gallon of milk (not “ultra-pasteurized”)
- 1.25 cups cool water (chlorine free)
- 1 teaspoon crystalline citric acid
- ¼ of a rennet tablet (1/4 teaspoon if using liquid rennet)
- 1 teaspoon cheese salt (Pickling salt) [You may choose to use a salt substitute and/or herbs]
- Baking soda (3 tablespoons)
- Vinegar (1/2 cup)

#### Equipment

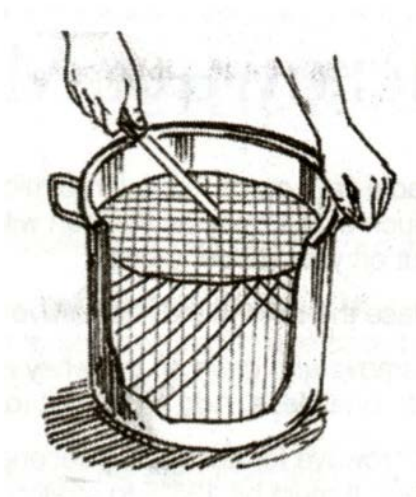
- 1 gallon stainless steel pot or non-aluminum cast iron pots.
- 3 half gallon stainless steel pots or non-aluminum cast iron pots.
- Thermometer
- Colander
- Slotted spoon
- Long knife
- Microwaveable bowl (if you want to use a microwave to heat curds)
- Rubber gloves (optional but recommended to help insulate your hands from hot curds)

Procedure:

1. Add 1 & 1/2 tsp. Citric Acid diluted in 1cup cool water to 1 gallon of cold milk.
  - a. If your lab group was designated the BASIC group, you will also add three tablespoons of baking soda.
  - b. If your lab group was designated the ACIDIC group, you will also add ½ cup mL of vinegar
  - c. If your lab group was designated UNALTERED group, you will not add anything.
2. Heat slowly to 90F. 90 degrees F = 32.2 degrees Celsius,
3. Remove pot from burner slowly stir 1/4 tab or 1/4tsp of rennet diluted in 1/4cup. cool water for 30sec. Cover and leave for 5 minutes.
4. Check the curd, it will look like custard and the whey will be clear. If too soft let set a few more minutes.



5. Now cut the curd into 1 inch squares with a knife that reaches the bottom of the pot.



6. Place pot back on stove and heat to 105 F while stirring slowly. (If you will be stretching in water heat to 110F). Note: 105 degrees F = 40 degrees Celsius.
7. Take off the burner and continue stirring slowly for 2-5 minutes. Transfer the curd to a colander or bowl using a slotted spoon.
  - a. Notice how the curd is beginning to get firmer as the whey drains.

8. Continue separating the curd and notice the color of the whey.
9. Drain the whey from the curd while gently pressing to aid whey runoff.



10. Using a heat proof bowl microwave on High for 1 min. pour off the whey.



11. Knead and reheat for 30 seconds, repeat if needed until the curd is 135F, almost too hot to handle.
12. Knead the curd in the bowl as you would bread dough.
13. Remove curd from bowl and continue kneading, return it to the microwave if needed.

14. At this point, if hot enough, it will begin to stretch. It is important to stretch it at least 20 cm.



- a. This step is important as it elongates the protein and causes them to bind with each other.

15. After stretching, knead the curd back into a ball.

16. When finished, submerge it in 50 deg. F water to cool for 5 minutes and then in ice water for 15 minutes. This will cool it down and allow the cheese to hold its shape. This step is critical as it protects the silky texture and keeps it from becoming grainy.

### Post Lab Questions

1. Did the unaltered cheese turn out? How did it taste?
2. How did raising the pH with baking soda influence the curd formation?
3. How did lowering the pH with vinegar influence the curd formation?
4. Why is it important to add the citric acid to the milk?
5. What do you think would happen if you boiled the milk, instead of just heating gently?