Blood Clotting to the Rescue: How to Stop Too Much Blood from Flowing

https://www.sciencebuddies.org/science-fair-projects/project-ideas/HumBio_p037/human-biology-health/blood-clotting (http://www.sciencebuddies.org/science-fair-projects/project-ideas/HumBio_p037/human-biology-health/blood-clotting)

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Experimental Procedure

Investigating Coagulation

In this part of the science project, you will make semisolid (gelatinous) balls using a solution of sodium alginate (dyed with food coloring) and a calcium chloride solution. This calcium chloride solution (which will have no anticoagulant in it) will be your **control** solution, meaning the one that should give clear, expected results and serve as a reference. The calcium in the calcium chloride solution should react with the sodium alginate to coagulate and form semisolid balls (made of calcium alginate), similar to the ones shown in Figure 1 in the Introduction (http://www.sciencebuddies.org/science-fair-projects/project-ideas/HumBio_p037/human-biology-health/blood-clotting#background). This coagulation process is similar to what happens in blood to form blood clots. To study the effect of anticoagulants on coagulation, you will test the effects of adding different amounts of sodium citrate (an anticoagulant) to the calcium chloride solutions. How do you think this will change your results? You will *quantify* your results by measuring the dimensions of the balls you make. *Note:* In this science project, we will refer to the calcium alginate products as "balls," but this does not mean they will necessarily be spherical.

1. As mentioned, you will be measuring the dimensions of the semisolid balls of sodium alginate to quantify your results. To do this, you should use graph paper with lines that are 1 millimeter (mm) or 2 mm apart. Then, when you later make the balls, you will place them on a sheet of graph paper to determine the balls' diameters. You will measure the balls' heights by using a cut-up piece of graph paper.
    a. To measure the balls' heights, cut off the edge of a sheet of graph paper so that the first lines of the grid are right at the edge of the paper. You could use part of the same sheet you are using to measure the balls' diameters, or you could use a new sheet. (You could cut out a strip of graph paper, making it like a ruler, if that is easier to work with.) Then, when you make the balls, you will place the cut-up graph paper behind the balls to determine their heights. For an example of how this should be done, see Figure 5.
    b. *Note:* The reason why you are cutting off the edge of the sheet of graph paper (and not using an uncut sheet of graph paper or an actual ruler) to measure the heights of the balls is because most graph paper and rulers have a little bit of space before the first marks. When you are making small measurements, this extra space could cause inaccurate measurements.
2. In your lab notebook, make a data table like Table 2. You will be recording your results in this data table.

		Diameter (mm)			Height (mm)	Observations
	Ball	**Longest diameter**	**Shortest diameter**	**Average**		
No sodium citrate	1					
	2					
	3					
	4					
	5					
	Average					
1% sodium citrate	1					
	2					
	3					
	4					
	5					
	Average					
1.5% sodium	1					

		Diameter (mm)			Height (mm)	Observations
	Ball	**Longest diameter**	**Shortest diameter**	**Average**		
citrate						
	2					
	3					
	4					
	5					
	Average					

Table 2. In your lab notebook, make a data table like this one. In it, you will record your semisolid ball measurement results. If no balls form, the diameter and height should be "0." *Note:* The "balls" may not be spherical.

3. Now make the sodium alginate solution.
    a. In the cup part of a blender, add 120 milliliters (mL) (1/2 cup [C.]) of cold tap water.
    b. Weigh out 2 grams (g) of sodium alginate and add that to the water in the cup.
        i. To weigh out the sodium alginate and other chemicals used in this science project, cut a small piece of wax paper (around 8 cm–10 cm on each side), place the wax paper on the scale, zero out the scale (so that it reads "0 g"), and then weigh out the chemical on the wax paper. Use a clean spoon to scoop the chemicals out of their containers. *Note:* You should use wax paper because it is harder for chemicals to stick to than normal paper.
        ii. *Tip:* If the scale you are using does not have a feature to zero it out, you will need to first weigh the piece of wax paper so that you can subtract this weight from the total when you weigh the chemicals on it.
    c. Add five drops of food coloring to the blender cup.
    d. Add another 120 mL (1/2 cup) of cold tap water to the cup.
        i. Adding the rest of the water now should help mix the sodium alginate and food coloring a little.
        ii. Your blender should now look similar to the one in Figure 2.

Figure 2. After adding the water, sodium alginate, and food coloring to the blender, it should look similar to the blender cup shown here. *Note:* The coloring of your mixture will be different if you did not use red food coloring.

e. You might want to ask an adult to help you use the blender to blend the water, sodium alginate, and food coloring so that the solution is fluid. When you are done blending, the solution should look like the one in Figure 3.
    i. Secure the blender cup lid tightly before blending so nothing gets spilled.
    ii. *Tip:* It may be easiest to make the solution fluid by blending the contents two or three times, for 5–10 seconds each time; if possible, shake the cup in between blendings.

Figure 3. After blending the water, sodium alginate, and food coloring together, you should have a solution that is *homogeneous* (all of the solution's parts are now mixed together and the solution looks the same throughout).

4. Next make the calcium chloride solutions. You will make three different calcium chloride solutions with different *concentrations* of sodium citrate (the anticoagulant).
    a. Set out three bowls.
    b. Label the three bowls using sticky notes or pieces of paper and tape.
        i. One bowl should be labeled *No sodium citrate*, another should be labeled *1% sodium citrate*, and the last should be labeled *1.5% sodium citrate*. *Note:* The solution with no sodium citrate added will be your control because it should give clear, expected results (in other words, semisolid balls should form).
    c. Add 240 mL (1 C.) of water to each bowl.
    d. Then add 1.3 g of calcium chloride to each bowl.
        i. Measure out the calcium chloride as you did in step 3b.
        ii. Be sure to use a fresh piece of wax paper so no leftover sodium alginate contaminates your solution.
    e. To the bowl labeled *1% sodium citrate*, add 2.4 g of sodium citrate.
        i. This is a 1% solution because 2.4 g divided by 240 mL equals 0.01, which equals 1% (0.01 multiplied by 100).
    f. To the bowl labeled *1.5% sodium citrate*, add 3.6 g of sodium citrate.

i. Can you figure out why this is a 1.5% solution?

g. Stir each bowl using a different, clean spoon until the calcium chloride and sodium citrate have completely dissolved.

5. Test if you can make sodium alginate balls using your different calcium chloride solutions. You should try to make a total of at least five balls with each particular solution, making and measuring only one ball at a time. Repeating your results ensures that they are robust and reproducible.
   a. Place a small piece of plastic wrap on a sheet of graph paper (for measuring the balls' diameters). Make sure the graph paper you prepared in step 1.a. (for measuring the balls' heights) is nearby.
   b. Make sure a timer, stopwatch, or clock that shows seconds is ready nearby.
   c. Using the syringe that came with the spherification kit, or a medicine dropper, suck up a small amount of the sodium alginate solution.
      i. If there is a layer of foam on the top of the solution, dip the syringe below that layer so you only suck up the liquid part.
      ii. If there is any foam or excess solution on the sides of the syringe, carefully wipe it off on the rim of the sodium alginate container.
   d. Practice releasing the sodium alginate solution very slowly back into its container. You want to get used to making one drop at a time.
   e. Once you can make one drop at a time, drop a single drop into the bowl containing the solution of calcium chloride without sodium citrate (your control solution).
      i. The tip of the syringe should be around 8–13 cm (3–5 inches) above the surface of the solution.
   f. Let the drop sit in the solution for 60 seconds (sec); the timing is important.
   g. After 60 sec, try to use a clean spoon to scoop the ball out of the solution, taking care to scoop as little of the solution out as possible without damaging the ball.
      i. If nothing clearly formed, make a note of this in your lab notebook. Do this by recording the diameter and height as a zero in your data table.
      ii. *Note:* When using the solution of calcium chloride without sodium citrate, a semisolid ball *should* form, so if it did not, you may want to re-check to make sure your solutions were prepared correctly.
   h. Measure the diameter of the ball by placing it on the plastic wrap on top of the graph paper and counting how many lines the ball spans. Note your findings in your data table.
      i. Move the plastic wrap around a little until the edge of the ball lines up with one of the lines, as shown in Figure 4.
      ii. Based on the number of lines the ball spans, calculate the ball's diameter in millimeters. If your graph paper has lines that are 2 mm apart, this means you will multiply the number of lines the ball spans by *two*.
         1. For example, in Figure 4, the ball spans about 2.5 lines on graph paper that has lines that are 2 mm apart. This means the ball has a diameter of 5 mm (since 2.5 times 2 mm equals 5 mm).
      iii. If the ball is not a sphere, measure its longest diameter and its shortest diameter and record both diameters in your data table. (Move the ball around by carefully moving the plastic wrap.) If the ball is a sphere, record the same number for both diameters. Record the diameters in millimeters.
      iv. *Tip:* If there is liquid around the ball, making it difficult to measure, you can carefully dab the liquid with a small piece of paper towel to remove the liquid. If you damage the ball when doing this, do not record the measurements for this ball and instead create a new ball in this solution (by repeating step 5.c.–5.h., and continuing from there). You may also want to record these observations in your lab notebook.

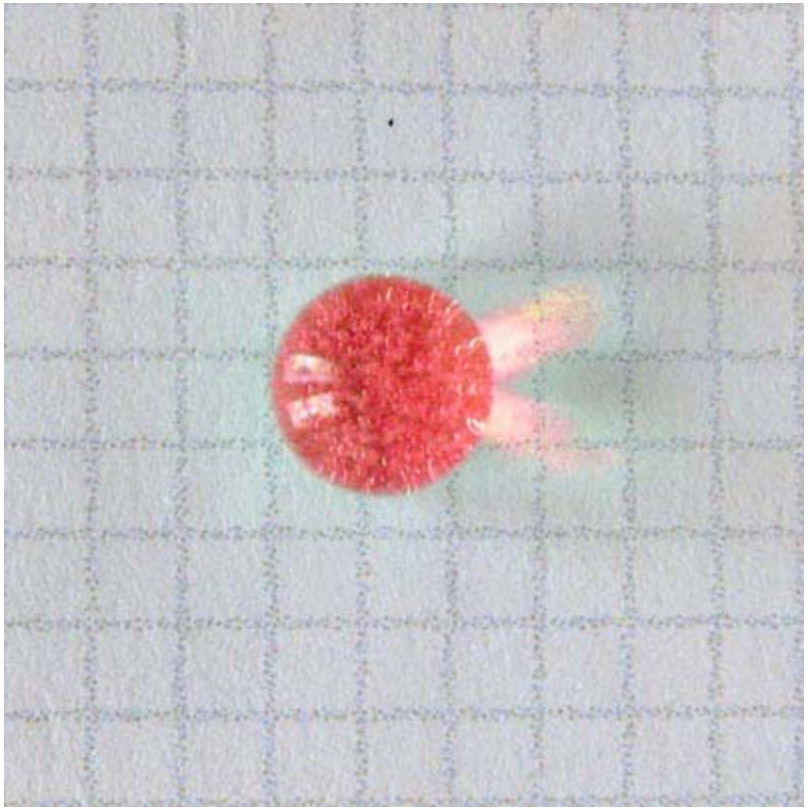

Figure 4. To measure the diameter of the ball, carefully move the plastic wrap so that one edge of the ball is lined up on a line, as shown here. Then count how many lines the ball spans. Measuring from the leftmost line, this ball spans about 2.5 lines, and since the lines in this graph paper are 2 mm apart, this ball has a diameter of about 5 mm (2.5 times 2 mm equals 5 mm).

i. Measure the height of the ball by placing the graph paper you prepared in step 1.a. behind the ball, as shown in Figure 5.
    i. Make your eye level with the ball and graph paper, such as by lowering your eye to the level of the counter or surface that you are using.
    ii. Record the height of your ball (in millimeters) in the data table in your lab notebook. Be sure to account for whether you are using graph paper with lines every 1 mm or every 2 mm.

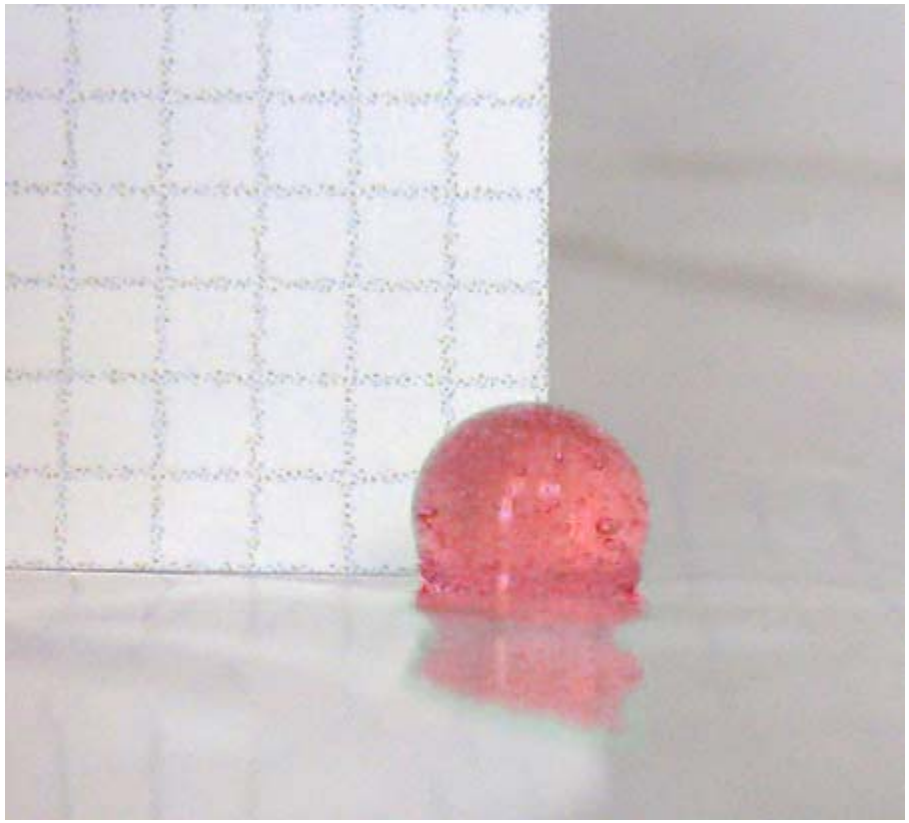

Figure 5. To measure the height of the ball, place the graph paper you prepared in step 1.a. behind the ball. Make your eye level with the ball and the graph paper and read how high the ball reaches on the graph paper. In this image, the ball and graph paper are not completely level (the viewer would need to lower his or her head to be level and make an accurate reading).

j. Repeat steps 5.c.–5.i. four more times so that you have made and measured a total of five balls using the solution of calcium chloride without sodium citrate.
    i. Make sure to record your data in the data table in your lab notebook.
k. Repeat steps 5.c.–5.j., but this time use the 1% sodium citrate solution.
    i. Try to make and measure at least five balls using this solution. Be sure to let each drop sit in the calcium chloride solution for 60 sec; the timing is important.
    ii. Make sure to record your data in the data table in your lab notebook.
l. Repeat steps 5.c.–5.j., but this time use the 1.5% sodium citrate solution.
    i. Try to make and measure at least five balls using this solution. Be sure to let each drop sit in the calcium chloride solution for 60 sec; the timing is important.
    ii. Make sure to record your data in the data table in your lab notebook.

6. When you are done testing, make some general observations about the balls. How do they look and feel compared to each other? Record your observations in

the data table in your lab notebook.
    a. If you have a camera, you may also want to take pictures of your results. You may want to take some from above and some from the side. Later, you could print your pictures and put them on your Project Display Board (http://www.sciencebuddies.org/science-fair-projects/science-fair/science-fair-project-display-boards).

Analyzing Your Data

In this part of the science project, you will analyze your data and come up with conclusions related to coagulation and blood clotting.

1. Look at the data table in your lab notebook and calculate the average diameter for all of the balls. Record these numbers in your data table.
    a. For example, if the widest diameter of a ball was 6 mm, and the shortest diameter was 5 mm, the average diameter would be 5.5 mm (since 6 mm plus 5 mm equals 11 mm, and 11 mm divided by two [the number of measurements] is 5.5 mm).
    b. If the widest and shortest diameters were the same (which would be the case if the ball was a sphere), the average diameter should equal these diameters.
2. Next, calculate the average ball diameter and height for each of the three different solutions. Record these numbers in your data table.
    a. For example, if the average diameters of the balls for one solution were 6 mm, 5.5 mm, 6 mm, 6 mm, and 6.5 mm, the average diameter would be 6 mm for that solution (since the sum of these numbers is 30 mm, and divided by five [the number of measurements] is 6 mm).
3. Make two bar graphs of your data: one graph of the average diameter for the balls made using the different solutions and one graph of the average height for the balls made using these solutions.
    a. You can make your graphs by hand or use a website like Create a Graph (http://nces.ed.gov/nceskids/CreateAGraph/default.aspx) to make the graphs on a computer and print them.
    b. For both graphs, put the solution names on the x-axis (the horizontal axis going across). Put either the average diameter or height (in mm) of the balls on the y-axis (the vertical axis going up and down).
        i. This means you should have three bars on each graph, one labeled *No sodium citrate*, one labeled *1% sodium citrate*, and one labeled *1.5% sodium citrate*.
4. Look at your data table, graphs, and observations and try to draw conclusions from your results.
    a. How did the balls change as more and more sodium citrate was added? Do you think your results indicate that the sodium citrate disrupted the coagulation process? Why?
        i. Did the balls' diameters and heights change in the same way (in other words, they both increased or decreased as more sodium citrate was added) or did they change in opposite ways (in other words, the width increased while the height decreased)? What does this tell you about how their overall shapes changed?
    b. What do your results tell you about how coagulation and anticoagulants work?
    c. Based on your results, why do you think coagulation is important in the blood clotting process? How do you think disrupting blood clotting can cause blood disorders?