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[CLICKING]

**SARAH**

All right. We can probably get started. So good afternoon. And today, we are going to start talking about the final lab that we are going to talk about in the lecture and the final lab that you haven't heard about yet for the course, which is the catalase lab. And some of you guys started that yesterday. And some of you will start it today. And then the rest of you will have to wait a little bit. But the catalase lab is our biochemistry type lab that we were doing in 5.310. And catalase is an enzyme if you couldn't tell by the name. A lot of enzyme names-- you guys are familiar from your biology classes-- end in ase.

**HEWETT:**

So catalase is an enzyme. And enzymes are proteins-- just a review of your general bio-- that carry out specific chemical reactions. And they're produced by different living organisms. They work in the cells to do a very wide range of chemistry. And there are many different graphical representations, weird pictures that you can find on the internet that try to depict what an enzyme actually does in cartoon form. And so here are a couple of examples of that.

So you have your enzyme and then you have what's called your substrate. And so that is the thing that the enzyme is doing chemistry on. The substrate comes in. It interacts with your enzyme to form an enzyme substrate complex. Then the chemistry happens. And then the enzyme releases the product. So sometimes the products will be rearranging the substrate. Sometimes it will be taking two substrates and joining them together. And sometimes it will be taking one substrate and breaking it apart. So there's a bunch of different reactions that can happen in different enzymes.

Similar thing-- if you want to look at your enzyme as a little Pacman kind of guy. You form the enzyme substrate complex. You form your products. And then they break apart. So that's the general idea of how enzymes works in a very quick nutshell. So our specific enzyme is catalase. And here are a couple of pictures of catalase or artist renditions of catalase. So this is a space filling model of what it would look like if all of the atoms were kind of 3D balls.

So you can see it has four different subdomains here. And then these darker parts in the middle with the little green dot, those are your heme center, so these are iron molecules and that's where the chemistry happens. So these are the active sites of the catalase. And you can also-- you may have seen in other courses proteins represented like this, which has a little bit more of the features of their secondary and tertiary structure. So you can see the alpha helices and beta sheets. And we're going to talk more about the structure of the active site and how the chemistry happens and what all of those squiggles mean next week when we talk about catalase.

**AUDIENCE:**

[INAUDIBLE]

**SARAH**

**HEWETT:**

Yes. Or wait. Each sub-domain has one active site. So it has one heme in each of the four parts. And we'll talk more about that next week when we go a little bit deeper into the structure of catalase and how it works. And then the other important thing you need to know about catalase is it's a large molecule. Proteins generally are. There are polypeptides-- lots and lots of amino acids. And so its weight is 240 kilodaltons. And a kilodalton is 1,000 mass units. So it's 240,000 grams per mole. And you'll need that for some calculations that you'll do in the second half of the lab. And we'll go over that again in the next lecture. But to give you an idea of the size of that thing and the molar mass, it is very large.

Why do we need catalase? So there are many redox reactions that are happening in your body where you do electron transfer reactions and you oxidize or reduce different things in your metabolism. And so electrons are transferred around cells. You may have heard of the molecule NADPH or NADH. Those transfer electrons between different molecules in your cells and help with metabolism. And while these electrons are being transferred around, you also know that we breathe oxygen and we need oxygen to do-- to perform many of our life processes. So you have a lot of oxygen in your body as well.

And during a lot of these metabolic processes you can get electrons that are transferred to oxygen. So if we have our oxygen and it gains an electron, we have what is called superoxide. So now this has a negative charge and an unpaired electron. And so it is a superoxide radical. And what do we know about radicals?

**AUDIENCE:**

[INAUDIBLE]

**SARAH**

**HEWETT:**

They would like to be stable. So they are not stable because they have this unpaired electrons. So they will either give up this electron or try to take an electron from something else in order to have their electron be-- or not be unpaired anymore. So these are very reactive in the body. And this is actually a problem. And we don't want this in our cells because it is so reactive. So there is another enzyme called superoxide dismutase that breaks this apart into elemental oxygen and oddly enough hydrogen peroxide. But it turns out that hydrogen peroxide in high enough concentrations is also toxic to your cells. And hydrogen peroxide can form hydroxy radicals, which are also very damaging to your cells.

There's a couple of reactions that hydrogen peroxide can do in the body. So the first is if you have hydrogen peroxide and then you have some of this superoxide around-- so it's a radical anion there. You can make oxygen, hydroxide ions, which are OK, and then hydroxy radicals. So then you form more radicals and those can go and damage your DNA, your proteins, the different fatty acids in your body, and cause all kinds of problems. Another thing that can happen is if there's just hydrogen peroxide around and it picks up an electron, it can form water and another hydroxyl radical.

So these are the reactions that we do not want to happen in our body because it forms these-- it just keeps propagating. And then when radicals react with other things that have all paired electrons, it generates more radicals and you propagate the radical formation throughout your cell it causes a lot of damage. So we need a way to get rid of the hydrogen peroxide in our body and that is what catalase is for.

And all organisms have catalase. You can extract it out of humans, all types of mammals, even plants have it. Cells do not want hydrogen peroxide in them. So there are different forms of catalase that you can get from any organism-- bacteria, all the things. The catalase that we're going to be using in the lab was extracted from cows. So just a fun fact.

And when we're talking about enzymes, the reason that enzymes are so good and so efficient at their jobs is because they catalyze chemical reactions. And a lot of you are chemical engineers, so you probably know about catalysis. So what does it mean for something to be a catalyst? Anyone? Please give me a quick catalyst definition.

**AUDIENCE:** [INAUDIBLE] reaction.

**SARAH**  
**HEWETT:** Yeah. It speeds up a chemical reaction. And it's not used up during the process. So if we look at our energy reaction diagram, if you have energy on this side and then reaction progress on this axis, you can say that you have reactants that are starting at a certain energy level. And then you have some energy that you have to put into the reaction to get it to go. And then in a lot of cases, if your reaction is exothermic, your products maybe at a lower energy level. So it'll release energy overall. But in order to get the reaction to go you have to put energy in. So this part of the curve is our  $\Delta H$  of our reaction. So it would be the energy released if it's exothermic, which in this case this reaction is.

But no matter what you need to put in some energy to get the reaction to go and this is called the activation energy. Occasionally abbreviated  $E_a$ . So this is what we care about when we're talking about reaction kinetics and what a catalyst does. So a catalyst functions and it makes the reaction faster by lowering this energy barrier. So if the yellow line is the reaction without a catalyst, then with a catalyst it will be smaller. And the products and the reactants will have the same total energy, but the energy you need to put in to get the reaction to go is going to be less.

And so the reaction can go faster and it can go in milder conditions. And we can talk about how enzymes work to lower this activation energy. So does anybody know what methods enzymes use to lower the activation energy of a reaction? Yes?

**AUDIENCE:** Proximity? Just getting the reactants together?

**SARAH**  
**HEWETT:** Yeah. Proximity. So it gets the reactants close together. If you have your reactants in a beaker and they're all floating around, they have to find each other before they can react. And so if you have an enzyme that has a specifically designed active site to hold these two molecules close together, then they will react. What else?

**AUDIENCE:** It can stabilize a transition state?

**SARAH**  
**HEWETT:** Yeah. It can stabilize a transition state. And how does it do that?

**AUDIENCE:** [INAUDIBLE]

**SARAH**  
**HEWETT:** Yeah. So it bonds to the substrate and it can have either electrostatic or even sometimes covalent interactions. And it stabilizes the transition state. And part of the way that it does that is it holds the molecules in the right orientation for them to react. So even if you have two molecules, if they're supposed to interact like this to react and they're in a beaker and they hit on this side or upside down, the reaction will not happen. So we can stabilize transition states. Your transition state and put molecules in the correct orientation. Anything else? There is one more thing that they can do. OK. What's your idea?

**AUDIENCE:** If they raise the temperature, it will increase-- collisions could increase.

**SARAH**

**HEWETT:**

So increasing the temperature of a reaction will get it to go faster. Right? But that's just because then you will have more molecules that have the necessary activation energy. So you're not necessarily lowering the activation energy that the reaction needs. But the last thing that they can do is they can provide alternate reaction pathways. So sometimes you may think that this will just be a one-step reaction. If you had an enzyme, occasionally it'll look like this. It might provide an extra intermediate in there that helps to lower the overall energy.

And so that's another way that it can lower this overall activation energy is by providing an alternate mechanism other than maybe just two molecules reacting together. There might be a third intermediate that helps actually lower the overall energy. So those are possible ways that the enzymes can work to lower the activation energy. And so our goals for the catalase lab are-- well, there are a few goals that we have. But our goals for days one and two are to determine the activation energy of hydrogen-- the decomposition of hydrogen peroxide as catalyzed by the catalase enzyme.

So we're going to try and figure out what the activation energy is after it's already been catalyzed. And in order to do this, we are going to need to first determine the order of the reaction with respect to hydrogen peroxide and be able to write a rate law. That'll help us characterize the kinetics. So in day one of this lab, you're going to do that first part. And you're going to figure out what the order of the reaction is. And in order to have that make any sense we need to talk about kinetics and how we discuss kinetics in a chemical sense.

So kinetics deals with the speed of chemical reactions. And if we use everybody's favorite generic reaction  $a + b \rightarrow c + d$ . You can write the rate of this reaction-- you can express it in a number of ways. So the first way is to think of the rate of the reaction as the change in the reactants over time-- so  $a$  and  $b$ . And it has a negative sign because you are losing the reactants. Hopefully they're going away as the reaction progresses. And it is equal to the rate of the appearance of  $c$  and the appearance of  $d$ . So these do not have negative signs because you are making your product.

So that is one way to express the rate of the reaction. And when you do it this way, you need to account for the fact that there are different molar ratios. So the reactants and the products will disappear in form at different rates depending on how many of them you need in the reaction. So to account for that, we multiply by the reciprocal of this coefficient. We can also write a generic rate law like this where your rate equals your rate constant, which you can calculate and determine in lab and is dependent on the temperature times the concentration of your reactants raised to a power and your concentration of your other reactants raised to a different power.

And you can also write an integrated rate law, which you may have seen before. So integrated rate laws deal with one reactant at a time. And so if we look at  $a$ , we can say-- we can write-- if there's only  $a$  we can write the rate of  $a$  equals the rate constant times  $a$ . Or we can say that the rate is equal to the disappearance of  $a$  over time. And you can take the derivative of this. So if we move everything over, rearrange it--

So if we integrate both sides of this-- if we integrate this side from your initial concentration to your concentration at some temperature and this side from  $t$  equals 0 to some time  $t$ , then if you do that math out you will get the natural log of-- this is your integrated rate law if your exponent here equals 1. So obviously, that will change if you have different exponents here. And then when you do your integration you will get different values.

But this is a way to relate the concentration at your initial concentration to a concentration at a final temperature or a final time. So that's one thing that you've probably seen before in some of your other chemistry classes. And we can also talk about-- well, we'll talk about that in a second. You may also have seen in some of your other classes enzyme kinetics or heard of Michaelis-Menten kinetics. And the Michaelis-Menten model of enzyme kinetics says that if you have an enzyme and you have a bunch of substrate, the reaction can only go as fast as there is when there's only one substrate in each enzyme.

So if all of the active sites are full of substrate, then that is as fast as the reaction can go. And it will proceed at a steady state. So you can make graphs of the reaction rate versus how much concentration of substrate you have. And you can get this Michaelis-Menten constant here, which is a measure of how much the enzyme-- what its affinity is for the substrate essentially. So does the enzyme need a whole bunch of substrate before it'll start going quickly? Or can it go quickly and efficiently with just a tiny concentration of substrate?

So we're not going to worry so much about those parameters for our lab right now. But this is something you may have seen. And one of the assumptions of the Michaelis-Menten kinetics, which is actually kind of important to us is that you have your enzyme and your substrate. They make the enzyme substrate complex. And then they go to enzyme and product. And they don't go in the reverse reaction. So that's kind of important to us that if we are trying to measure the rates of our reactions, that we don't have to contend with the reactions being catalyzed in reverse. All right.

So now we can take a little bit of a closer look at the rate law for just a normal chemical reaction. And pretty much the most important thing you want to know about this is that the units of the rate are always moles per liter per second. So whenever you are trying to calculate a rate constant, you want your units to line up. So if your rate-- if we want this in molarity per second-- if your rate equals-- let's say these are both equal to 1 just for now. Then what are our units over here? M and M. So what are the units of k have to be?

So that's how you can figure out what the units of your rate constant are because it always needs to multiply with your concentrations and your k to equal moles per liter per second. And that is going to be the units of our rate that we care about in our kinetic equations. The rate constant is different at different temperatures. So if you change the temperature of the reaction, then your rate constant will change and you'll have to remeasure that.

And x and y are values that need to be determined. They may be the same as little a and little b but not always. So the kinetics of a reaction are determined if it's a multi-step reaction if their reaction mechanism has more than one part in it. They may not all be reflected in this overall chemical equation. You may not see them, but the rate is going to be dependent on your slowest step. And that slowest step is what is going to determine your rate law. And it may not always match your overall reaction equation. So be careful when you are determining these things. You can't always just assume that it is from the little a and little b.

So how are we going to determine the order for our reaction? And this is what are going to do on day one. We will use our rate law expression to determine the order of the reaction. So you're going to-- according to our rate law here. It's the only reactant that we have in this case is hydrogen peroxide, and we need to determine what its order is.

So if we change the concentration of the peroxide, we should change the rate, yes? So if we do a bunch of reactions with a bunch of different concentrations of peroxide, we can measure the rates, and then use the relationship between those two things to figure out our concentration of-- or our order of the reaction.

So the way that you're going to do this in lab is you will have a series of solutions, and these are your solutions. You'll have a stock hydrogen peroxide solution that you're going to make up. It's going to be about 4% hydrogen peroxide. You'll use a phosphate buffer. Why do we need to use a buffer for this reaction? What's a buffer, I guess is the first question?

**AUDIENCE:** It helps the solution from changing PH2.

**SARAH HEWETT:** Perfect. So a buffer helps the solution, it prevents a solution from changing PH. So you can make a buffer out of certain PH, and then if acid or base gets added to it, it resists the change in PH. It keeps the constant. Why is that important for our reaction?

**AUDIENCE:** Maybe [INAUDIBLE].

**SARAH HEWETT:** Yeah. So enzymes are most functional at a set PH. So your PH of your body is around 7, so we want to keep the PH of our reaction somewhere around our physiological PH, so our buffer is going to be set to PH 6.8, which is going to help make sure that the enzyme is in its active state. It is not denatured. If you change the PH too much, you'll change the protonation state of the different parts of the protein, and it can fall apart.

So we want to make sure that this is happening at the correct PH. We'll add some enzyme, and then we will measure the rate of the reaction. And we're going to do that using this apparatus right here. So you'll have one of these. You'll have a water bath, so that we can keep the temperature constant. So you'll just fill this with room temperature water, and then you can monitor the temperature throughout your reaction to make sure that it does not change.

And then this is your reaction vessel here, your pressure tube, because you will be putting all of your solutions in here. You will add your peroxide and your buffer, and then you will put this cap on. And this Teflon cap here, it's important to note, has an O-ring on the bottom. And so you want to make sure when you're doing this reaction that you're O-ring is in there, because that'll make a good seal, so you won't have your products escaping because our product is a gas.

So you'll screw this on. And then there's a hole in the top here. You can inject your enzyme really fast. You're going to be doing this in partners. Somebody can inject the enzyme really fast. And then you want to connect your pressure tube. And this tube is connected to a pressure sensor. And this pressure sensor will be connected to your computer, which is why you need to bring a laptop for the catalase lab.

And we will give you the Logger Pro software. And it will send the data directly to your laptop. And it'll plot it for you. And you'll get a curve that, hopefully, looks something like this. And we will measure the pressure of oxygen formed in kilopascals versus time in seconds. And it'll graph this thing for you.

And the slope of the graph is going to be your rate of kilopascals per second. And we want to take the rate at this first part of the graph where it is linear, and why do we want to take the rate at the beginning of our reaction?  
Yes.

**AUDIENCE:** Reaction rate is dependent on amount of reactants?

**SARAH**

**HEWETT:**

Yeah. So the reaction rate is dependent on the amount of reactants, and the-- so the rate of this reaction obviously changes over time, and we set up our reactions so that we know the concentration of peroxide. And the only time in this reaction that we're theoretically going to know the concentration of peroxide is at the beginning, before some of it has reacted and the rate changes. So we want to measure the initial rate so that we know our concentration, and that we get a-- we have a consistent place to take our data from on each measurement.

So once we have done all of these different reactions-- you'll do all four of them-- all of the same procedure in the same tube-- you'll make five different graphs. You'll have this. Then how do we analyze this data and get information about the order out of it? So here we are going to have to do a little bit of math.

So here are some of the steps of the data analysis. The first thing that you're going to have to do is calculate the concentration of the peroxide that you used. And we are going to give you 30% hydrogen peroxide, which is-- its volume per volume-- so there's 30 milliliters of H<sub>2</sub>O<sub>2</sub>, and 70 milliliters of water.

So if you remember that your molarity is your moles of your solute over the total volume of the solution, you can turn this milliliters, assuming a density of about 1, into grams, grams to moles, and you can calculate the molarity of this solution. You're then going to take this 30% hydrogen peroxide that we give you, and dilute it 13.3 milliliters to 100 milliliters. So then you can do another dilution equation to figure out your final concentration of your hydrogen peroxide that you're going to be using in the lab.

So that's a first step. And then I'm going to kind of rearrange a couple of these other steps, just because it'll make this a little bit easier to go through. But the first thing that we're going to do is the easy part, which is get your rate of oxygen formation in kilopascals per second. And that we're just going to read straight from the slope of the graph. So that's pretty easy. So we'll have our rate of oxygen formation in kilopascals per second. Great.

So now we want to take a look at our rate law. And we have our rate of oxygen formation, but what's in our rate law? Hydrogen peroxide. So we need to find a way to go from oxygen and kilopascals per second to moles per liter per second of hydrogen peroxide.

So the first way that we're going to do that is to go from kilopascals per second here to molarity of oxygen. And we can do that using our gas constant here. So if you divide by the gas constant, multiply by 1 over 8.314 moles, liters times kilopascals, we can cancel out our kilopascals.

And we'll have moles per liter, which is good, but then we still have this K, which is our temperature in Kelvin, so we also have to divide by our temperature in Kelvin. And this will get us our rate of oxygen in moles per liter per second, which is pretty good. We're close. But we still need to get from oxygen to hydrogen peroxide. So how can we get from oxygen to hydrogen peroxide using this information?

**AUDIENCE:**

[INAUDIBLE]

**SARAH**

It would be-- so the ratio is 2:1, right. So there's two hydrogen peroxides in this reaction for every one oxygen.

**HEWETT:**

But these coefficients are in moles. And this is in moles per liter. So we need to figure out how many moles of oxygen we have before we can use our stoichiometry ratio here.

So how are we going to figure out how many moles of oxygen we have? We need to know what our volume of oxygen was. And for that we can go to our pressure tube. So you'll have some of this is going to be your solution down here, so that'll be liquid. And then we need to calculate the volume of the gas above our reaction, which is where oxygen is going to be formed.

So we'll need to know the volume of this, and the volume of the pressure tube. And the volume of the pressure tube has already been measured, and we'll give that to you. And you guys are going to measure in the lab what the volume of your reaction vessel is. You're going to fill this thing with water, weigh it, then weigh it empty, and then you can calculate by difference in the density of water what your volume of your whole tube is.

And then you know your reaction volume is 25 milliliters, so you can subtract those and get the volume of the gas. So if we know these two together, you'll get the volume of the gas above your reaction. So if we have our rate of oxygen formation in moles per liter per second, we can multiply that by our volume that we calculate, so liters of oxygen. And then we have our rate in moles of oxygen per second.

And now we can use our stoichiometry, because we have it in moles, so we can take our rate in moles per second, and then we know that it is 1 mole of oxygen for 2 moles  $\text{H}_2\text{O}_2$ . Then we get our rate  $\text{H}_2\text{O}_2$  in moles per second. And what are the units of rate that we want? Moles per liter per second.

So we have it in moles per second. We're so close. Then we just need to divide by the liters of our reaction, which if you go back a couple of slides, is 25 milliliters, so all of our reaction volumes are 25 milliliters. So if you convert that to liters, divide by 0.025 liters, and then we will have our rate in moles per liter per second, which is what we want.

Now this still doesn't get us to a, but we can rearrange this equation by taking the natural log of both sides. So if you have the natural log of the rate equals the natural log of K plus, then the A comes down, a times the natural log of your hydrogen peroxide concentration, then-- we've just figured this out from all of that math. We know this concentration.

And this is going to be our y-intercept. So if we have  $y = b + mx$ , if we make a graph of the hydrogen-- or the natural log of the peroxide concentration and the natural log of the rate, then our slope is going to be our order. Hopefully, we haven't lost you.

So the way that you're going to treat all this data is you can, instead of having to do this calculation out for every single trial, you can make a giant spreadsheet. And in the lab manual, it tells you what data we want you to collect. And this is all of it here. And then you can make a spreadsheet that does all of the calculations for you, that gets you down to the natural log of rate, and the natural log of hydrogen peroxide.

So this is an example of a pretty good way to present your data. And I know it's kind of hard to see on the printed out slides, but these will be on Stellar if you want to look at them. And then your graph will look something like this. And you'll have a straight line. And so from that straight line, you can figure out what your order of your reaction is based on your slope. So that was a lot.

So once we have that done on day one, then, if you remember our second goal for the lab, it was to figure out the activation energy of the reaction. And so the way that we're going to do that is to use the Arrhenius equation, and that says that the rate constant is equal to A, which is this collision frequency factor, times e to the negative activation energy over RT.



And if we remember the things that can change the rate, we can change the concentration and that'll affect the rate, and also the temperature. So if we do our reaction at different temperatures, then we can determine our activation energy. So what you're going to do is you're going to pick one of the trials from day one. Usually people pick the middle one because this is easy. It's just 1 milliliter, no decimals. Just easy to measure and it's right in the middle.

You'll pick one and you'll hold the concentration of peroxide constant, and then we will vary the temperature. So you'll do seven runs at varying temperatures. So you'll pick a temperature in each of these ranges and you will measure the rate. Again, you'll get the same graph, the measure the rate of oxygen per second. And then you can do all of this same math to get your information in concentration of peroxide to go from oxygen to peroxide, because that's what we care about.

The keys to success for this reaction are to wait for the peroxide and the buffer to reach the correct temperature before you add the enzyme. So when you have your reaction here in this tube, you'll put it in your water bath and then you want to give it a few minutes to stir and come to equilibrium at the right temperature, because it'll all be at room temperature. And then if you're trying to do your reaction at 0 degrees or 5 degrees, your reaction part won't actually be at 5 degrees even if your water bath is. So give it a few minutes to equilibrate before you take your measurements and before you inject your enzyme.

Another key to success is to start at the hottest temperature and then add ice to cool it. It's easier to control the cooling than it is to control the heating with these hot plates. The hot plates tend to get really excited and then they'll heat your solution up way above what it needs to, and then it won't be able to cool. So if you start at your hottest temperature and then add ice, you'll be able to slowly lower the temperature and do it in a more controlled fashion.

The other important thing about this reaction for day one and two, and I'm sure your TAs will emphasize this when they tell you in lab and you guys have a chance to have this in front of you, is to do everything really quickly. So the reaction starts as soon as you add the enzyme. So you'll have your micropipette, you'll stick it right in that hole, add the enzyme really fast, and then you want to attach your pressure tube and then hit go on your computer to start collecting the data. And that all has to happen in like, a second so that you don't miss that beginning of your data collection. So those are some important things.

If we talk about the data analysis for this reaction, it's kind of similar to the other one. So you'll calculate the concentration of peroxide, you'll calculate the volume of air above your reaction. You'll have the initial rate of oxygen formation in kilopascals per second. Then you can get the initial rate of oxygen formation in molarity per second and moles per second. Then you can change your rate of oxygen formation to the rate of peroxide decomposition, same math.

And then using the rate law that you determined on day one, you're going to calculate the rate constant. So if we have our rate, our general rate law, we'll measure this in the lab or we'll calculate this, I guess. We will know this from day one.

So you'll plug in the value that you get from your first set of calculations right there. And then we measure this in the lab or we can calculate/measure this. So we can calculate our rate constant, which is, if you just rearrange this, it'll be  $k$  -- so that's not so bad.

And then in order to get our activation energy out of this, we are again going to take the natural log of both sides. I think that I did that out correctly. No plus sign here.

All right, so this is your linear form of this equation. So we have, again,  $y$  equals  $b$   $m$  and  $x$ . So if we do our reaction at different temperatures, then we will know the temperature of our reaction. And then we will get  $k$  from our rate constant from our rate law, and you'll calculate that for each of your trials, as well, based on the rate.

And then our slope is going to be the activation energy over the ideal gas constant. So we can get the activation energy from the slope of a graph of  $1$  over  $t$  times the natural log of  $k$  versus the natural log of  $k$ . And then you can also get your collision frequency factor, this  $A$  term, from your  $y$ -intercept, so you'll also be able to determine that.

Then you'll use your Linus program to get the errors in all of these things, and that is essentially day one and day two of the lab. Do you guys have any questions about that? There's a lot of math.

So again, there's another data chart that you can make for day two of the lab that has all of the information that you're going to need for these calculations. And then this is hopefully what your graph will look like. It should be linear with a negative slope, and then the negative will get taken care of with that negative sign in the equation.

**AUDIENCE:** This will be posted on Stellar?

**SARAH**  
**HEWETT:** Yes. Yeah, all of the slides will be posted on Stellar so you can see it in a larger form, because I know they didn't print out super well.

OK. So now we can take a look at the reaction that you're going to do in a slightly different form. And you may have seen this before, but we're going to do it anyway. So hydrogen peroxide, you may have some at your house. They sell it in a drugstore. It's like, 3% so it's not as concentrated as the stuff that we're going to be using in the lab. But nonetheless, hydrogen peroxide is a thing that you can buy.

And it naturally decomposes over time, so this reaction is always happening. If you have a bottle of hydrogen peroxide, usually the caps are vented because if you seal it for too long, you'll build up pressure of oxygen and that's a problem. The stuff that we have in the lab is 30%, so that's definitely vented, and we store it in the refrigerator so that we slow down the reaction and keep our product for longer.

So it is always happening and you can speed it up in a number of ways. So one of the ways is obviously using enzymes, catalase. That'll speed up the reaction. But there are other catalysts that you can use, as well.

So have you guys seen the elephant toothpaste reaction before? Yes. Do you guys know what the catalyst is that we use for this? There's a few you can use, but does anyone remember?

So we're going to react hydrogen peroxide with potassium iodide in this case. And you can use a couple different ones, like manganese dioxide, I believe, works. You can use different iodide salts, but potassium iodide is the one that I have today. And we're going to do a little bit of an experiment.

So we said that concentration affects rate, right? So we will try our first reaction with some hydrogen peroxide. And I'm going to pour out about 10 milliliters of this and then dilute it so that our new concentration is about 7%. Yeah, that's right. So pour that in there.

And then we want to use a potassium iodide solution. So we can pour some of this in here. And then we have the oxygen bubbles, and who did the catalase reaction yesterday in lab? Anybody who's here?

So what did you guys see when you added the enzyme to your peroxide in the tube? Bubbles, good. But it didn't foam up, right? Because they just popped and formed oxygen.

So the way that we can get this to foam is by adding some soap. This is just powdered soap. And we can add a couple of scoops of that. And now hopefully, if all goes well, when I add these together, we will see the decomposition of hydrogen peroxide.

I can probably hold it up so you can see what's happening. So what's happening in there? It's bubbling, good. Excellent. So what are the bubbles? Oxygen. All right, so it's taking its time.

So while that is going, we can set up another one. And this time, we won't dilute the hydrogen peroxide, so we'll just use 40 milliliters of hydrogen peroxide. Need our soap.

Yeah. Yeah, so it looks like toothpaste maybe. Toothpaste that an elephant would use, yeah. What? Elephant toothpaste. Yeah.

I also, in the course of doing some research for this, found that some people call it old foamy, like a geyser. So you may have heard it by that name.

All right, and then we add the same amount of potassium iodide. Hopefully I measured this out properly. So if that was 7% hydrogen peroxide, what do we think is going to happen if we use 30%? Faster. Let's find out.

So can you see the steam coming off of it? Yeah, so what does that mean about the reaction? It's exothermic. Yeah, it's very hot. If you were to come up here and touch this, it is quite warm.

But yeah, so that went faster, right? So we can do a little bit of math about how much faster the potassium iodide makes the reaction go than just what would happen in your bottle at home. And so our activation energy of hydrogen peroxide decomposition that is uncatalyzed, just happens in nature, is 75 kilojoules per mole.

So if we use our Arrhenius equation, then we can plug in 75 kilojoules or 75,000 joules to match our or gas constant units. And then the activation energy of the decomposition as catalyzed by potassium iodide is about 56 kilojoules per mole, so it lowers it, makes the reaction go faster. If you do this math out-- I don't want to get my numbers wrong here-- it is about 2,500 times faster or 2,400 times faster than what would happen in nature.

Now, if we talk about catalase, and you guys will calculate the actual activation energy of the catalase-catalyzed reaction, I will start and tell you that it is less than 10 kilojoules per mole. So that is a pretty significant change, right? Going from 75 kilojoules per mole down to 10 or less. That's a lot.

So if you do this math out, where you go from 75,000 joules to 10,000 joules, does anyone have any guesses as to how much faster the catalase is than just normal decomposition? A lot.

So that's a lot faster. So you can be thinking about that when you are thinking about what happens in the cells of your body because you are forming these superoxide radicals and hydrogen peroxide all the time in your cells. And this reaction is happening on this scale, or the reaction that you just saw is happening this many times faster in your body, and you will see it happen also in your flask.

So any questions, thoughts? No? All right, well, thank you guys. You can head to lab.