EZ BioResearch Bacteria Science Kit (10-Pack)

(Pre-poured LB Agar Plates and Cotton Swabs)

EZ Science Fair Project E-Book 2015 V-3

www.ezbioresearch.com Tel: 1(800) 637-0262 Fax: 1(877) 693-4868

About EZ Science Fair Project E-Book

In this EZ Science Fair Project E-Book, we designed ten sets of experiments exploring various aspects of bacteria and their interaction with us. The first four sets of experiments (experiments #1, #2, #3 and #4) will show that we are surrounded by bacteria. Bacteria are present everywhere, in our home where we live, in our school where we study, on personal objects which we touch and use, and even on our own body parts that we take in our food and air. These experiments will help children/students to visually understand that bacteria are present in our surrounding although we cannot see them when bacteria are present in single or small numbers of cells. The second two sets of experiments (experiments #5 and #6) will allow us to measure the effectiveness of our cleaning methods, which will help children/students to establish good personal hygiene. The next two sets of experiments (experiments #7 and #8) will explore the fruit that we eat and the famous 5-second rule. Experiment #9 demonstrates that some bacteria are beneficial to our life. The last experiment #10 will show children/students how to quantitatively measure and compare the effectiveness of various antibacterial agents.

These ten experiments with different variables can be easily expanded to about fifty or more experiments. Hopefully these proposed experiments can provide you with some guide lines and some stimulating ideas about designing award-winning science fair projects.

We plan to add more experiments to our EZ Science Fair Project E-Book. Here is a link for you to check and download any future update to the EZ Science Fair Project E-Book.

http://www.ezbioresearch.com/E-Book2015v2_ep_54-1.html

If you have any question or have trouble in downloading the E-Book, please feel free to contact us at (800) 637-0262 or support@ezbioresearch.com.

Enjoy and have fun!

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About EZ BioResearch LLC

EZ BioResearch is an innovation-driven biotech company which focuses on developing, manufacturing and distributing advanced molecular biology tool kits and high quality laboratory consumable products. (www.ezbioresearch.com)

EZ BioResearch offers microbiology tools and reagents including bacteria science kits, bacteria DNA isolation kits, agar plates, antibacterial agar plates, culture tubes, Petri dishes, inoculating loops and needles, T-spreaders, etc. All our bacteria science kits are manufactured and packaged in US by EZ BioResearch. All products and kits have gone through strict quality control procedure to ensure high quality of the products.

EZ BioResearch Bacteria Science Kit has been ranked #1 best selling product since 2012 in pre-poured agar plate category. EZ BioResearch Bacteria Science Kit received more than 300 reviews, ten times more than other vendors. 99% of our customers are satisfied with their purchases. Many customers won science fair project competitions using EZ BioResearch Bacteria Science Kit. We are encouraged by the overwhelming number of the good reviews that we received and we are also grateful for those customers with constructive feedback. We actually improve the quality of our products by adding new desired features requested by our customers.

EZ BioResearch is proud to be your partner in your research and committed to deliver the most innovative products with the highest quality. We continually improve and expand our product portfolio to include the best products in the market. We have established a comprehensive quality assurance program to meet and exceed the expectation of our customers. We guarantee that the products that we offer are the best possible quality for the price.

Contact Information:

If you have any technical question, please call us at 1(800) 637-0262 or email us at **support@ezbioresearch.com.**

If you have any question or problem on your Amazon prime membership order, please call Amazon Customer Support directly at 1-866-216-1072, 24 hours a day, 7 days a week.

If you have any question or problem on your Amazon non-prime membership order, please call us at 1(800) 637-0262 or email us at **support@ezbioresearch.com.**

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Welcome and Congratulations!

Thank you for purchasing our #1 best selling EZ BioResearch Bacteria Science Kit! At EZ BioResearch, we would like to congratulate you on making the right choice. There are many reasons that you may choose EZ BioResearch Bacteria Science Kit over others. However, we would like to point out to you two most critical advantages of purchasing EZ BioResearch Bacteria Science Kit that you may or may not be aware of.

1) **SAFE FOR CHILDREN/STUDENTS:** Our Bacteria Science Kit is safe for children/students. Children/students' safety is of our utmost concern when we design bacteria science kits. We use Luria Broth (LB) based medium in our agar plates. LB medium is a nutrient rich medium which allows fast and proliferative growth of bacteria. LB is the most commonly used medium in the research laboratories all over the world. It is safe for children/students to use due to its non-selectivity. Not all bacteria growth media are safe for children/students to use. Tryptic Soy (TS) medium, one of the other known nutrient media on the market, is not safe for children/students to use. Due to its selectivity, it may selectively produce or enrich harmful pathogens which may cause illness to the children/students. You may check the science buddies website (http://www.sciencebuddies.org/science-fair-projects/project_ideas/MicroBio_Agar.shtml) to get more safety information about the bacterial culture medium. Many parents and teachers who purchased Tryptic Soy agar plates might not be aware of the danger that they brought to their children/students. Please do spread the words to other students/parents/teachers and educate them about the potential danger. Doing science experiments is fun but safety of our children/students is more important.

2) **FREE TELEPHONE TECHNICAL SUPPORT:** You not only purchased Amazon #1 best selling bacteria science kit, but also gained free access to professional technical supports from EZ BioResearch. For many students/parents, this may be your first science fair project and also your first time looking into the invisible micro-world. You may have many questions that need immediate answers. We are the only company in Amazon that offer live telephone support on bacteria science kits. The reason is quite simple. We are a true biotech company with a physical address. We have well trained technical stuff on call during the working hours. Some of other vendors on Amazon do not have a physical address or a telephone number. They only have a PO Box or UPS Box to collect checks. Where do you think they manufacture the agar plates? Probably in the kitchen or garage! Please do spread the words to other students/parents/teachers that not all science kits are made alike and inform them about the free telephone technical support from EZ BioResearch.

At EZ BioResearch, we are dedicated to make learning science easy and fun. We put together this EZ science fair project E-book to help you gain some basic knowledge of microbiology and provide you a few stimulating ideas about science fair projects. When you finish your science fair projects, please go to Amazon and share your thoughts and your science fair projects with us. Please do upload some pictures of your experiments. We love to see pictures! Have fun!

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Introduction

Description: EZ BioResearch Bacteria Science Kit (Pre-poured LB Agar Plates and Cotton Swabs) is a convenient and ready to use science fair project kit designed to help children/students learn microbiology. LB stands for Luria Broth, a nutritionally rich medium, is one of the most common medium used in the laboratories worldwide. LB medium is safe for student use. This Kit is great for school science fair project competition. No messy microwave or heating is needed. Each kit is backed with our 100% money back guarantee.

Kit Contents:

Components	Quantity
LB Agar Plate	10 plates
Cotton Swab	10 swabs in 5 pouches
Hand Sanitizer	1 bottle
Transfer Pipette	2 Pipettes
Dilution Tube	2 tubes

Storage: All our agar plates are produced in our sterile manufacturing facility and go through our stringent quality control inspection. The agar plates can be stored at room temperature for 30 days. However, whenever possible, try to store them in a refrigerator. Low temperature will keep the agar plates fresh, minimizing the loss of the water from the surface of the agar plates. Agar plates stored under such conditions have a minimum shelf life of 3

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months. **Do not put the agar plates in a freezer!**

Technical Support: Any question? No worry. You will receive immediate live support with PhD scientists with extensive hands-on experience. Call us at 1-800-637-0262 or email us at support@ezbioresearch.com

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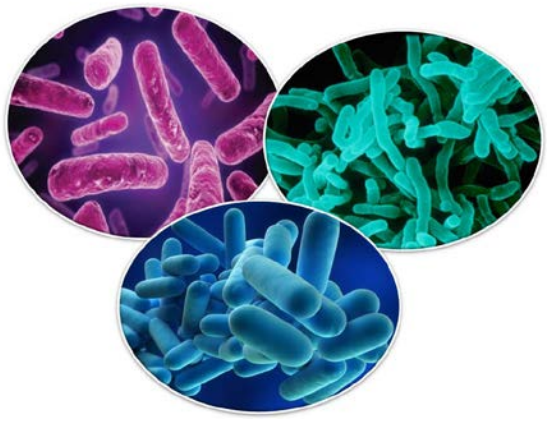
Background Information on Bacteria



- Bacteria are everywhere. Bacteria play all kinds of roles in the environment and living organisms. There are good bacteria and bad bacteria. Good bacteria can help enrich nutrients in the soil, help us properly digest food and absorb nutrients. Certain good bacteria are essential for decomposition of waste materials. People also utilize the good bacteria to make food such as cheese, yogurt and fermented foods. Bad bacteria can attack plants, animals and human. Bad bacteria attack organisms by releasing toxic chemicals that are poisonous to plants and animals, thus cause illness.

- Bacteria are single celled organisms. They are so small that are invisible to our naked eyes. In order to see bacteria, a single bacteria cell needs to grow and replicate by about 1 million times. The best medium to grow bacteria is called Luria Broth (LB) medium. LB medium is the most commonly used nutrient-rich medium and it is safe for children/students to use.

- Bacteria need proper temperatures to grow. At low temperatures (<70^{o}F), most bacteria either stop growing or grow very slowly. At high temperature (>100^{o}F), many bacteria will not grow and will die due to excessive heat. Therefore, for science experiments, it is important to set up an incubator of 85-100^{o}F so that the growth of the bacteria can be easily seen. A simple incubator can be made of a plastic storage box with a clamp/desk lamp or portable heater. Use a thermometer to make sure that the temperature is between 85 and 100^{o}F before placing the agar plates in the incubator. Take temperature measurements over a couple of hours until the temperature is steady. **Do not let the temperature exceed over 100^{o}F. When using agar plates to culture bacteria, do not blow air directly over the agar plates as it will dry out the agar plates**.

- Bacteria need water to grow. Bacteria are present everywhere. However, in dry places, bacteria are few and mostly in a dormant state, whereas in the moist environment, more and active growing bacteria are mostly seen.

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General Experimental Procedure

1) Take out one sterile cotton swab. Wipe a test object with the cotton swab to pick up bacteria. Here are some of the popular test objects: human or pet teeth, hands, phone, TV remote, restaurant menu, sink, floor, fruit, vegetable etc. If the test object is dry, the cotton swab needs to be dampened with bottled water.

2) Take out one of the agar plates (No warm-up is needed). Open the agar plate and GENTLY wipe the agar plate with the cotton swab to transfer the bacteria from the cotton swab onto the surface of the agar plate.

3) Cover the agar plate and place it in an upside-down position in an incubator with a temperature between 85 and 100 ^{o}F. A desk lamp or a portable heater can be used as a heat source.

4) Watch appearance of small bacterial colonies on the surface of the agar plates after 12-48 hr of incubation. Take pictures of the agar plates to keep them as a record.

5) Terminate the experiment after 5 days. Put the used agar plates in a zipper-lock bag and dispose of them as regular trash. Prolonged incubation is strongly discouraged due to possible contamination in a non-sterile environment.

Most Frequently Asked Questions:

1) Why are there no bacteria after 3 days at room temperature? The most likely reason is that the room temperature is too low (below 75 ^{o}F). An incubator with a temperature between 85 and 100 ^{o}F is strongly encouraged.

2) I do have an incubator. Why is there no bacterial growth? The most likely reason is that the temperature is too high (>100 ^{o}F) during the incubation. Even for a few minutes above 100 ^{o}F, the heat will kill the bacteria. A thermometer is needed to measure the temperature and to make sure that the temperature in the incubator is stable. It takes one to two hours for the temperature in the incubator to reach equilibrium. Also the thermometer should be placed near the agar plates closest to the heating source. The other possibility is that the test object is dry. You need to prewet the cotton swab to pick up the bacteria.

3) Why are all my agar plates dried out within two days? The most likely reason of loss of moisture in the agar layer is due to either rapid air circulation (use a fan) or higher incubation temperature (heater).

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Experiment #1
Question: Where can you find bacteria inside your home?

We know that bacteria are present everywhere. However, bacteria are so small that are invisible to our naked eyes. According to some research, there are six dirty places in our home. They are kitchen sink, toothbrush, salt and pepper shaker, TV remote control, computer keyboard and bathtub. Surprisingly, the toilet is not on the list. You can use the kit to confirm/challenge the research results or test your own questions.

Experiment Procedure:

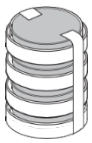
1. Label agar plates on the bottom (not the lid) with the object names that you want to test.
   Note: *To save material and test more objects, you may use a marker pen to draw a line at the bottom of the plate to divide one plate into two sections.*

2. Take out one sterile cotton swab. Wipe a test object with the cotton swab to pick up bacteria.
   Note: *If the test object is dry, the cotton swab needs to be dampened with bottled water.*

3. Take out one of the agar plates (No warm-up is needed). Open the agar plate and GENTLY wipe the agar plate with the cotton swab to transfer the bacteria from the cotton swab onto the surface of the agar plate.

4. Repeat steps 2-3 until you finish all the objects you would like to test.
   Note: *use new cotton swab for each test object.*

5. Follow the General Experimental Procedure on page 6 to grow bacteria.
   Note: *Do not forget to invert the agar plates before putting them in an incubator.*



6. Watch appearance of bacterial colonies on the surface of the agar plates after 12-48 hr of incubation.

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7. Count the bacterial colonies on each agar plate and determine which object is the dirtiest object.

You may test some dry surface objects such as carpet floor, table, chair, etc, and test some wet surface objects such as inner rim of the toilet, kitchen sink, bathtub, etc. Compare the result to see which have more bacteria. Make sure that the collection surface areas are about the same size.

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Experiment #2
Question: Where can you find bacteria in your school?

According the survey conducted by National Sanitation Foundation International, bacteria are commonly found in the following school areas: water fountain spigot, reusable plastic cafeteria tray, faucet, keyboard, toilet seat and student hands. Use this kit to test where you can find bacteria in your school and see which objects have the most bacteria.

Experiment Procedure:

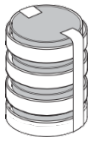
1. Label agar plates on the bottom (not the lid) with the object names that you want to test.
   Note: *To save material and test more objects, you may use a marker pen to draw a line at the bottom of the plate to divide one plate into two sections.*

2. Go around your school and find the objects you want to test. Wipe the test objects with the cotton swabs to pick up bacteria. Use a separate cotton swab for each object.
   Note: *If the test object is dry, the cotton swab needs to be dampened with bottled water.*

3. Take out one of the agar plates (No warm-up is needed). Open the agar plate and GENTLY wipe the agar plate with the cotton swab to transfer the bacteria from the cotton swab onto the surface of the agar plate.

4. Repeat steps 2-3 until you finish all the objects you would like to test.

5. Follow the General Experimental Procedure on page 6 to grow bacteria.
   Note: *Do not forget to invert the agar plates before putting them in an incubator.*



6. Watch appearance of bacterial or fungal colonies on the surface of the agar plates after 12-48 hr of incubation.

7. Count the bacterial colonies on each agar plate and determine which object is the dirtiest object.

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Experiment #3
Question: What is the dirtiest object that you have?

There are many objects that you may touch, play, and/or use daily. As a student, you may use pencils, pencil sharpener, rulers, erasers, books, desk, computer keyboard, iPhone, iPad, favorite toys, sports equipment etc. How clean are they? Which one is the dirtiest? Let us find out.

Experiment Procedure:
Since this is a comparison among all the objects, it is only fair that the test surface areas are of the same size. An area of 2 x 2 inches on each of the object should be enough for this purpose.

1. Make a list of the objects that you choose to compare and label the agar plates on the bottom (not the lid).
   Note: *To save material and test more objects, you may use a marker pen to draw a line at the bottom of the plate to divide one plate into two sections.*

2. Select a test area of 2 x 2 inches on each of the objects and swab the area with cotton swab to collect bacteria.
   Note: *If the test object is dry, the cotton swab needs to be dampened with bottled water.*

3. Follow the General Experimental Procedure on page 6 to grow bacteria.

4. Watch appearance of bacterial colonies on the surface of the agar plates after 12-48 hr of incubation.

5. Count the bacterial colonies on each agar plate. Make a graph and compare to see which object has the most number of bacterial colonies.

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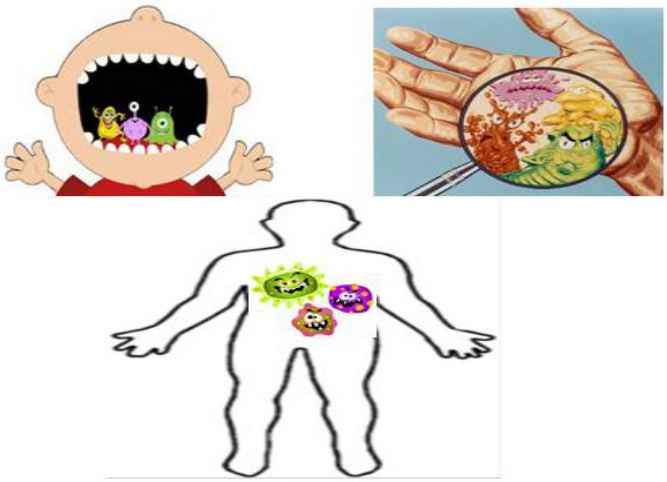
Experiment #4
Question: Are there any bacteria on your body?



If you have done the first set of experiments #1-#3, you must have known that bacteria are all around us. But are there any bacteria on our body? You may say yes because our hands touch those objects that contain bacteria. What about other parts of our body? For example, our lips, our teeth, our tong, our nose, our hair, our arms, our arm pits and of course our smelly toes? Here is a simple experiment that will tell you the answer.

Experiment Procedure:

1. Make a list of the body parts that you will test and label each of the agar plates on the bottom. Please do include the teeth. You will know the reason after you get the result of this experiment.

2. Select a test area of approximately the same size (2 x 2 inches) for each test subject and swab the area with cotton swab to collect bacteria.
   Note: *If the test object is dry, the cotton swab needs to be dampened with bottled water. For teeth, just use one cotton swab rubbing the teeth area near the gum (the area where plague is usually located). Cover 4 or 5 teeth and rub back and forth 4-5 times. The best time to swab your teeth is either in the morning before brushing the teeth or 1 hrs after meal.*

3. Follow the General Experimental Procedure on page 6 to grow bacteria.

4. Watch appearance of bacterial colonies on the surface of the agar plates after 12-48 hr of incubation.

5. Count the bacterial colonies on each agar plate and see which part of your body has the most number of bacterial colonies.

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Experiment #5
Question: What is the best way to clean our hands?

There are several ways that we can use to clean our hands, rinsing with water, using soap, or rubbing with a few drops of hand sanitizer. Are they all equally effective to remove bacteria from our hands? There are several variables in the experiment that we can change and see the effect of these variables. As to rinse with water, how long do we have to rinse it? Rinse for 5 seconds, 15 seconds or 60 seconds? Is warm water better than cold water? How about rubbing hands vs. without rubbing hands? As to the soap, are different brands of soaps equally effective in making our hands clean? As to the hand sanitizer, how many drops are needed?

Experiment Procedure:
In all the previous set of experiments (#1-#4), we tested different objects or places. The key is that the collection surface is about the same size. In this and next experiments, we will test the same objects with different treatments. **The key is to have a pretreatment control for each treatment.** For example, you may select one finger to test the presence of bacteria before washing hands. Then wash your hands and select another finger to test the presence of bacteria after washing hands. (The reason to use two different fingers in the experiment is that we may remove some of the bacteria after we wipe it with the cotton swab. So it is better to use different fingers before and after treatment.) Here is how we calculate the effectiveness of cleaning. Let us say before washing, there are 100 bacteria colonies and after washing, there are only 15 bacteria colonies left. Thus the effectiveness of the washing is 85%, (100-15)/100. You can do the same calculation when using soap washing. Comparing the percentages will give you a fair comparison between two cleaning methods.

Be sure that your hands have roughly the same dirtiness for each experiment. You may play a ball in the yard or play with lawn grass for a few minutes before each experiment so that you get roughly the similar amount of bacteria on your hands.

Here is a simple experiment to compare rinsing with water vs. washing with soap.

1. Label 4 agar plates on the bottom: *before rinsing with water, after rinsing with water,*

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before washing with soap, after washing with soap.

2. **For rinse with water experiment:** Play a ball in the yard for 1 minute. Collect bacteria on the index finger on the left hand using a pre-wetted cotton swab (wet with bottled water). Then wash your hands with water for 5 seconds. Dry your hands with paper tower. Then collect bacteria on the middle finger on the left hand using a new pre-wetted cotton swab.

3. **For wash with soap experiment:** Play a ball in the yard for 1 minute. Collect bacteria on the index finger on the left hand using a pre-wetted cotton swab (wet with bottled water). Then wash your hands with soap. Dry your hands with paper tower. Then collect bacteria on the middle finger on the left hand using a new pre-wetted cotton swab.

4. Follow the General Experimental Procedure on page 6 to grow bacteria.

5. Count the bacterial colonies and calculate the percentage of bacteria removal (the effectiveness of cleaning) as described above. Compare the two methods.

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Experiment #6
Question: What is the best way to clean our teeth?

Ever since we are able to hold a tooth brush, we are taught to brush our teeth every morning and/or before going to bed. If we do not, what happens? We have a smelly morning breath. Yuck! Smelly morning breath tells you that you have a lot of bacteria on your teeth. There are many ways to clean up your teeth. We can rinse with water, brush with tooth brush and tooth paste, rinse with mouth wash, chew a chewing gum, clean with a WaterPik and with an electronic tooth brush, etc. Which method do you regularly use? Does your tooth cleaning method remove all the bacteria? Which method is the best way to clean your teeth? Which brand of the tooth paste is better than the others? With this bacteria science kit, you can get the answer yourself!

Experiment Procedure:
Similar to Experiment #5, we need to measure bacteria before and after treatment. Then we compare the percentage of bacterial removal for each method or condition.

Here is a simple experiment that you can use. Let us compare three methods to clean your teeth: rinse with water (day 1), brush with tooth paste (day 2), rinse with mouth wash (day 3).

The best time to do this experiment is in the morning when you just get out of bed. You need three days to complete the experiment, one cleaning method per day. Try to eat the same type of food for dinner for three days so that you will hopefully have the similar amount of bacteria on the teeth in the morning.

1. On day 1, we will test the effectiveness of rinse with water in cleaning teeth. Label 2 agar plates on the bottom: *before water rinse, after water rinse.*
   Note: *To save material and test more objects, you may use a marker pen to draw a line at the bottom of the plate to divide one plate into two sections. One section labeled as before and the other section labeled as after water rinse.*

2. Use one cotton swab rubbing the teeth area near the gum (the area where plague is usually located). Cover 4 or 5 teeth and rub back and forth 4-5 times.

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3. Transfer the bacteria from the cotton swab to the agar plate labeled as *before water rinse*.

4. Rinse your mouth with water. Repeat the rinse two more times.

5. Then repeat Step 2 and 3 to collect bacteria and transfer bacteria to the agar plate labeled as *after water rinse*.

6. Follow the General Experimental Procedure on page 6 to grow bacteria.

7. Count the bacterial colonies and calculate the percentage of bacteria removal (the effectiveness of cleaning) as described in Experiment #5.

8. On days 2 and 3, perform the same experiment by using tooth paste and mouth wash. Compare the results of the three methods and see which method is the most effective way to clean up your teeth.

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Experiment #7
Question: Are there any bacteria on fruit?

Among all the foods that we eat, we often eat fruit raw. Some of the fruit such as orange, we peel off the skin before we eat. Others like strawberry and blueberry, we eat them with skin on. Of course, we wash them before eating them. Does the skin of the fruit that we eat contain bacteria? Are there any bacteria under the skin of the fruit? How effective is our washing method? We can do some experiments to get the answer.

Experiment Procedure:

1. First, select 4 kinds of fruit for this experiment, strawberry, peach, apple and orange.

2. Label 8 agar plates on the bottom: *strawberry outside, strawberry inside, peach outside, peach inside, apple outside, apple inside, orange outside and orange inside.*

3. Follow the General Experimental Procedure on page 6 to collect and grow bacteria.
   Note: *If the surface is dry, you need to prewet the cotton swab with bottled water. Make sure that the collection surface area is about the same size.*

4. Count the bacteria colonies on all 8 agar plates and compare!

Which fruit has the most number of bacteria? If you find one, then you can wash the fruit and then test how effective your washing method is. You may vary different washing parameters such as soak time, number of rinse, with or without detergent, warm water vs. cold water. Following the similar experimental design as illustrated in Experiment # 5 and #6.

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Experiment #8
Question: Is 5-second rule a fact or a fiction?

You must have heard the famous five-second rule, right? When a food is dropped on the floor, it will not be significantly contaminated with bacteria if it is picked up within five seconds of being dropped. Is that really true? For this type of experiment, there are several variables (conditions) that we can change. One variable is the type of floor: hardwood floor, carpet floor, ceramic floor, outside floor vs. inside floor. Another variable is the type of food: dry food such as bread and cookies vs. wet food such as a slice of a tomato, a slice of apple, and a slice of ham. Another viable is the time that we leave the food on the floor: 1 second, 5 second, 1 minute, 5 minutes or several hours.

Experiment Procedure:
Similar to Experiments #4 and #5, five seconds of food touching the floor is considered as a treatment. Therefore, we need to collect bacteria on the food before the treatment, and collect bacteria on the food after the treatment. Different from Experiments #4 and #5, we expect to see an increase of bacteria on the food after the treatment.

Here is a simple experimental design to compare dropping of a slice of ham on the kitchen floor vs. on the floor outside of the front door.

1. Label 6 agar plates: *1) ham, 2) ham on kitchen floor, 3) kitchen floor, 4) ham, 5) ham on outside floor, 6) outside floor.*

2. Select the same size on the floor and ham. (2 x 2 inches).

3. Use a pre-wetted cotton swab to pick up bacteria on the ham slice and transfer the bacteria to the agar plate labeled as ham.

4. Then drop the ham slice and leave it on the floor for exactly 5 seconds. Pick up the ham slice and collect the bacteria on the surface of the ham which touches the kitchen floor. Transfer the bacteria on the agar plate labeled as ham on kitchen floor.

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5. Collect bacteria on the kitchen floor and transfer to the agar plate labeled kitchen floor.
6. Repeat the same experimental steps 3 to 6 using ham and the outside floor.
7. Follow the General Experimental Procedure on page 6 to grow bacteria.
8. Count the bacterial colonies on each agar plate and compare the numbers.

Is the five-second rule still valid? You may also vary the different conditions of the treatment as described above (time, floor type, food type).

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Experiment #9
Question: Are there any good bacteria?

Despite the fact that many of the bacteria are pathogen and associated with diseases, there are also many beneficial bacteria. The beneficial bacteria help us maintain the natural balance of organisms in the intestine, which is important for proper food digestion and immune functions. People also utilize good bacteria to manufacture certain food products such as yogurt, cheese, sour cream, kimchi and pickles. Because of the importance of beneficial bacteria in keeping us healthy, food manufacturers often add live beneficial bacteria to certain dairy products such as yogurt. You may notice that many yogurt products have labels of "contain live/active cultures" on the container. This means there are live beneficial bacteria in the yogurt. Some of the beneficial bacteria species you can find in yogurt are *Lactobacillus bulgaricus, Lactobacillus acidophilus, Streptococcus thermophiles,* etc. Since the bacteria are very small, it is hard for us to detect them by naked eye. With the EZ Bacteria Science Kit, you may test if there are live bacteria in the yogurt that you eat.

Experiment Procedure:

1. Obtain a cup of fresh, un-opened yogurt labeled as containing live and active cultures such as Activia.
   Note: Make sure the yogurt is within the best before date. We have tested different brands of yogurts and found Activia gives the most consistent result.

2. You will use the quadrant streak technique to inoculate the yogurt on the agar plate (see the illustration diagram below). Use a new cotton swab to pick up small amount yogurt and transfer the yogurt to the agar plate by gently rubbing the cotton swab back and forth about five times in the top left corner of the plate.
   Note: Make sure not to break the agar surface.

3. Turn the plate 45^{o} counterclockwise and using a new cotton swab touch the edge of the area that you just streaked, extend the streaks into the second quarter of the plate by rubbing the cotton swab back and forth about five times.
   Note: Avoid passing the swab into the first streaking area during back and forth rubbing.

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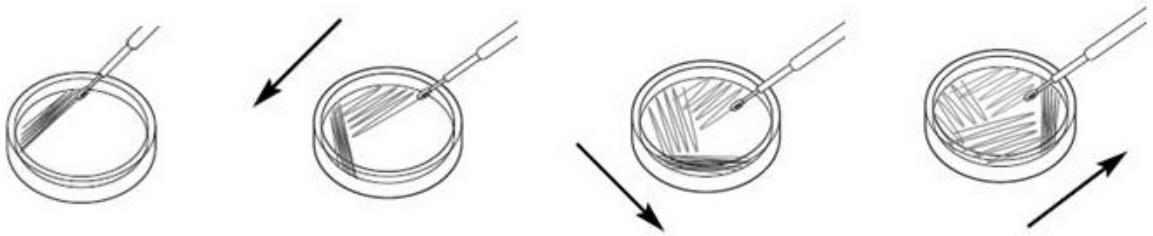
4. Turn the plate 45° counterclockwise and using the same cotton swab touch the edge of the second streaks, extend the streaks into the third quarter of the plate by rubbing the cotton swab back and forth about five times.
   Note: Avoid passing the swab into the first and second streaking areas during back and forth rubbing.

5. Turn the plate 45° counterclockwise and using the same cotton swab touch the edge of the third streaks, extend the streaks into the fourth quarter of the plate by rubbing the cotton swab back and forth about five times.
   Note: Avoid passing the swab into all previous streaking areas during back and forth rubbing.

6. Follow the General Experimental Procedure on page 6 to grow bacteria.

Are there any bacteria colonies on the plate? Do these colonies look different in terms of size, shape and color?

Quadrant Streak Technique



Step 1 Step 2 Step 3 Step 4

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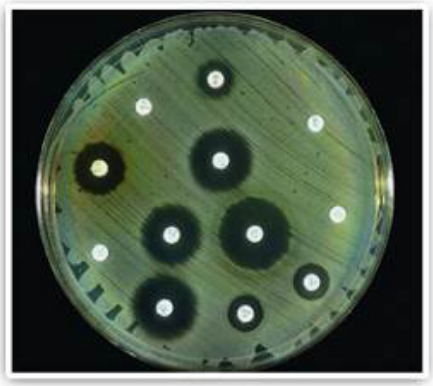
Experiment #10
Question: What is the best anti-bacterial agent?



In this experiment we will use the Kirby-Bauer disk diffusion method to measure the effectiveness of anti-bacterial agents. With this method, a "lawn of bacteria" must be first made on the agar plate by swabbing the bacteria of interest evenly across the plate. Then the filter paper disks bearing different antibacterial agents are placed down on the surface of the agar. The antibacterial agents will diffuse from the disks into the agar. If an antibacterial agent is effective in stopping the bacteria from growing, there will be a clear circular area around the disk (see above picture). The clear circular area is called the zone of inhibition. The large the clear area is, the more effective the antibacterial agent is. By comparing the diameters of the clear area, we can tell which one is the most effective anti-bacterial agent in the test.

Experiment Procedure:
You may test the effectiveness of antibacterial agents on various bacteria. For this particular experiment, we will test to see which mouthwash is the most effective one in stopping the growth of bacteria from your mouth.

1. On day 1, use one cotton swab rubbing the teeth area near the gum (the area where plague is usually located). Cover 4 or 5 teeth and rub back and forth 4-5 times.
   Note: *The best time to do this experiment is in the morning when you just get out of bed.*

2. Take out one of the agar plates (No warm-up is needed). Open the agar plate and GENTLY wipe the agar plate with the cotton swab to transfer the bacteria from the cotton swab onto the surface of the agar plate.

3. Follow the General Experimental Procedure on page 6 to grow bacteria.
   Note: *Do not forget to invert the agar plates before putting them in an incubator.*

4. On day 2, after 12-24 hr of incubation, you should see the bacteria colonies on the agar plates.

5. Make several disks using a hole punch on filter paper (coffee filter paper will work). Use a pencil label each disk with a code for each mouthwash you want to test. Wrap the disks

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with aluminum foil and put in a 300°F oven for 30 min to sterilize the disks. After 30 min, let the disk cool to temperature.

6. Fill one of the sterile dilution tubes included in the kit with clean bottled water to half full (~2 ml). Use a clean toothpick, pick up 2-3 bacteria colonies from the agar plate and place the colonies into the 2 ml water. Use a cotton swab to disburse the colonies into the water.
   Note: *Make sure that the colonies are evenly disbursed.*

7. After the bacteria colonies are disbursed into the water, take the cotton swab out of the bacterial solution and make sure to remove excess of water by gently pressing the cotton swab again the wall of the dilution tube. Wipe the wetted cotton swab gently across the entire surface of a new agar plate.
   Note: *You may need to dip the cotton swab in the bacterial solution one more time to complete the process.*

8. Cover the plate and let it sit at room temperature for about 5 min.

9. Use the transfer pipette included in the kit to soak the filter paper disk with different mouthwash you want to test. Remember to rinse the transfer pipette with clean bottle water in between each mouthwash. Do not forget to include a disk soaked with bottle water as a negative control.

10. Use a clean forceps to pick up a single disk, drain off excessive liquid and gently place it down on the surface of the agar plate that you just swabbed. Repeat the process until you finish all the disks. Make sure that you give enough space between each disk.

11. Follow the General Experimental Procedure on page 6 to grow bacteria.

12. Measure the diameter of the zone of inhibition for each disk and determine which mouthwash is the most effective one in killing the bacteria from your teeth.

13. To quantify the effectiveness of antibacterial agents, you can measure the diameters of the clear zone and rank the effectiveness of antibacterial agents by the diameters of the clear zone. You can make a table and a chart as well as a picture panel to show different effectiveness of antibacterial agents.

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