



MICROBE HUNTER™ Series

SURFACE MICROBE HUNTER

Code 5561

TEACHER GUIDE EXCERPT

This is a condensed version of the Teacher Guide for the Surface Microbe Hunter. Content for all sections is not included but section titles are listed to show the organization of this product.

Surface Microbe Hunter - Product Description

The Surface Microbe Hunter uses the exploration of the microbes found on surfaces to teach students STEM-based skills that they will apply to five classroom activities.

The SURFACE MICROBE HUNTER (5561) contains:

- TSA/RB BioPaddles®, 10 (Code 5552)
- Hand lenses, 10
- Surface Microbe Hunter™ CD
- Link to free BioPaddles® Colony ID™ Lite App

NOTE: The activities are set up for 10 student groups. Activity 1, and the follow up – Activity 2, require a total of 10 paddles. Activity 3, Activity 4, and Activity 5 each require 10 paddles.

The Surface Microbe Hunter CD contains the Student Guide and Teacher Guide which include background materials and instructions for each of the five activities. The Teacher Guide also includes recommended topics for student review and answers to student questions. Resources for the activities - including additional activities, and PowerPoint presentations on related topics– are provided on the CD. Digital files can be viewed directly by the students or files can be printed and distributed.

The BioPaddles® Colony ID™ Lite app (included) and BioPaddles® Colony ID™ App for iPads aid students in the presumptive identification of microbe growth on BioPaddles® with a large library of microbe images, ID guides, and resource materials. A report containing a full color image from an iPad camera can be emailed directly from the BioPaddles® Colony ID™ app. For more information go to the App Store.

CONTENTS

Standards, Objectives, and Requirements

STEM Education Standards Overview

Surface Microbe Hunter Objectives

STEM Extension Objectives

Do Antimicrobials Keep Products Cleaner?

How Clean Are Kitchen Sponges?

Are Spore-Forming Microbes Affected by Disinfectants?

Build an Incubator

Time Requirements

Equipment Requirements

Organization of the Surface Microbe Hunter

CD Contents

Safety

Surface Microbe Hunter

Surface Microbe Overview

Surface Microbes

Surface Characteristics

Activities

ACTIVITY 1 Are all Surfaces Alike? (Introductory to Intermediate Structured Activity)

ACTIVITY 2 Identifying Surface Microbes (Intermediate Structured Activity)

ACTIVITY 3 How Do Sanitizers Work on Surfaces? (Intermediate to Advanced Structured Activity)

ACTIVITY 4 Design a Sanitizing Protocol -
ENGINEERING ACTIVITY (Advanced Open Activity)

ACTIVITY 5 Antimicrobial Surfaces (Introductory to Intermediate Open Activity)

References

Resources

STANDARDS, OBJECTIVES, AND REQUIREMENTS

Stem Education Standards Overview

Skill / Concept	SCIENCE	TECHNOLOGY	ENGINEERING	MATH
<p>Experimental / Engineering Design</p> <p>Investigating</p> <p>Scientific Method</p> <p>Measurement</p> <p>Data Analysis</p> <p>Communication</p> <p>Technology</p>	<p>A.1.2 Design and conduct scientific investigations.</p> <p>A.1.3 Use technology and mathematics to improve investigations and communications.</p> <p>A.2.1 Conceptual principles and knowledge guide scientific inquiries.</p> <p>A.2.3 Scientists rely on technology to enhance the gathering and manipulation of data. New techniques and tools provide new evidence to guide inquiry and new methods to gather data, thereby contributing to the advance of science. The accuracy and precision of the data, and therefore the quality of the exploration, depends on the technology used.</p> <p>E.1.1 Identify a problem or design an opportunity.</p> <p>E.1.3 Implement a proposed solution.</p> <p>E.1.4 Evaluate a proposed solution.</p> <p>E.1.5 Communicate.</p>	<p>2.AA Identification of the criteria and constraints of a product or system.</p> <p>8.H Begin the design process</p> <p>9.K Create a prototype to test a design concept.</p> <p>11.O Refine the design.</p> <p>11.P Evaluate the design solution.</p> <p>11.R Communicate observations.</p> <p>12.O Operate the system to validate the design.</p>	<p>ET1 (Designed World) Study of designed systems, processes, materials, and products.</p> <p>ET1.A (Products, Processes, Systems)</p> <p>ET1.B (Nature of Technology)</p> <p>ET1.C Using Tools and materials)</p> <p>ET 2 (Engineering Design) Creative and iterative process for identifying and solving problems under constraints.</p> <p>ET2.A (Defining and Researching Technical Problems)</p> <p>ET2.B (Generating and Evaluating Solutions)</p> <p>ET2.C (Optimizing and making Tradeoffs)</p> <p>ET3 (Technological Systems) Effectively using technology systems.</p> <p>ET3.A Identifying and Modeling Technological systems)</p> <p>ET3.C (Control and Feedback)</p> <p>ET4 (Interactions of technology & Society) Decisions are affected by technology.</p> <p>ET4.A (Interactions of technology & society)</p> <p>ET4.B (Interactions of Technology and Environment)</p> <p>ET4.C (Analyzing issues involving Technology & Society)</p>	<p>1.0 Understand numbers, ways of representing numbers, relationships among numbers, and number systems.</p> <p>2.0 Algebra: Understand numbers, ways of representing numbers, relationships among numbers, and number systems.</p> <p>3.0 Geometry: Analyze characteristics and properties of two- and three-dimensional geometric shapes and develop mathematical arguments about geometric relationships.</p> <p>4.0 Measurement: Understand measurable attributes of objects and the units, systems, and processes of measurement.</p> <p>5.0 Data Analysis & Probability: Formulate questions that can be addressed with data and collect, organize, and display relevant data to answer them.</p> <p>6.0 Problem Solving: Build new mathematical knowledge through problem solving.</p> <p>7.0 Recognize reasoning and proof as fundamental aspects of mathematics.</p> <p>8.0 Organize and consolidate their mathematical thinking through communication.</p> <p>9.0 Connections; Understand how mathematical ideas interconnect and build on one another to produce a coherent whole.</p> <p>10.0 Create and use representations to organize, record, and communicate mathematical ideas.</p>
<p>Concept Principles & Knowledge</p>	<p>Biofilms</p> <p>Biodiversity</p> <p>Biodiversity Index</p>	<p>Cleanliness</p> <p>Cleaners, sanitizers, disinfectants</p>	<p>Data analysis; constructing tables and graphs</p> <p>Surfaces (surface types)</p>	
<p>Consolidated STEM Standards</p>	<p>S = National Science Education Standards (NSES) - K-4, 5-8, 9-12</p> <p>T = International Technology & Engineering Educators Association (ITEA) - K-2, 3-5, 6-8, 9-12</p> <p>A framework for K-12 Science Education: Practices, Crosscutting Concepts, and Core Ideas (NRC; 2011) - Draft</p> <p>E = Accreditation Board for Engineering and Technology (ABET) - 11-12</p> <p>A framework for K-12 Science Education: Practices, Crosscutting Concepts, and Core Ideas (NRC; 2011) - Draft</p> <p>M = National Council of Teachers of Mathematics (NCTM) - PreK-2, 3-5, 6-8, 9-12 Consolidated STEM Standards</p>			

SURFACE MICROBE HUNTER OBJECTIVES – STUDENTS WILL:

ACTIVITY 1 Are All Surfaces Alike?

- Form hypotheses about surfaces and microbes.
- Sample four major surface types
- Evaluate microhabitats
- Use BioPaddles to confirm their hypotheses
- Enumerate surface microbes
- Calculate a microbial diversity index for each surface type

ACTIVITY 2 Identify Surface Microbes

- Characterize and identify surface microbes in an incubated sample

ACTIVITY 3 How Do Sanitizers Work On Surfaces?

- Learn about the effectiveness of cleaning
- Create biofilms
- Evaluate the effectiveness of a surface sanitizer
- Compare the effectiveness of surface sanitizers
- Compare the effectiveness of cleaning procedures

ACTIVITY 4 Design a Sanitizing Protocol

- Design a glassware sanitizing protocol for a three compartment sink
- Evaluate sanitizing protocols for the cleaning and sanitizing of glassware.
- Compare the effectiveness of sanitizing protocols

ACTIVITY 5 Antimicrobial Surfaces

- Design a sampling method to evaluate the effectiveness of brass, stainless steel or other materials as an antimicrobial surface for door hardware such as doorknobs and push plates.
- Compare the antimicrobial effectiveness of materials used for high touch surfaces.

STEM EXTENSION ACTIVITY OBJECTIVES – STUDENTS WILL:

Do Antimicrobials Keep Products Cleaner? – Students will:

- Form a hypothesis about advertising claims regarding cleanliness and antimicrobial products
- Design an experiment to investigate the effectiveness of a commercial antimicrobial in mouse pads
- Compare diversity and microbe counts in products with and without antimicrobial additives

How Clean Are Kitchen Sponges? - Students will:

- Determine if sponges are a favorable surface for microbe growth.
- Investigates the difference in total colony count between a dry sponge vs. a damp kitchen sponge
- Investigates the role detergent plays in reducing microbe growth

Are Spore-Forming Microbes Affected by Disinfectants? – Students will:

- Design an experiment that evaluates the effectiveness of certain sanitizers on microbial spores in commercial paper strips impregnated with spores of *Bacillus stearothermophilus*.
- Compare the contact impact of selected sanitizers on spore-forming microbes such as *B. stearothermophilus*.

Build an Incubator – Students will:

- Design an incubator that will produce a constant temperature
- Investigate settings and configurations to determine which combinations will produce constant, specified temperatures for the desired length of time

TIME REQUIREMENTS

ACTIVITY 1	Are all Surfaces Alike? (Introductory to Intermediate) 45 minutes; 5-7 day incubation period; 2 periods
ACTIVITY 2	Identify Surface Microbes (Intermediate) 45 minutes
ACTIVITY 3	How Do Sanitizers Work on Surfaces? (Intermediate to Advanced) 45 minutes; 5-7 day incubation period; 2 periods Teacher must prepare samples and microscope slides prior to the activity; 2 hours of growing and drying time – not hands on.
ACTIVITY 4	Design a Sanitizing Protocol (Advanced – Engineering Activity) 45 minutes; 5-7 day incubation period; 2 periods
ACTIVITY 5	Antimicrobial Surfaces (Introductory to Intermediate) 45 minutes; 5-7 day incubation period; 2 periods

EQUIPMENT REQUIREMENTS

Incubator	An incubator set at 35°C (98°F) is preferable, although microbial growth will occur at lower temperatures, it will take longer – at least 5 days. If you do not have an incubator, incubate in the warmest part of a room, or place BioPaddles® on top of a refrigerator. Warm air from the coils is a favorable incubation environment. Alternatively, have students follow the STEM Build an Incubator activity to build an incubator. Expect identifiable colonies after 18 – 24 hours at 35°C (98°F) or after 5 – 7 days at 20°C (68°F).
Sink	Bleach solution used to disinfect colonized paddles can be poured down the drain with large amounts of running water and adequate ventilation.
iPad and App	The BioPaddles® Colony ID™ Lite app and BioPaddles® Colony ID™ app for iPads aid students in the presumptive identification of microbe growth on BioPaddles® with a large library of microbe images, ID guides, and resource materials. A report containing a full color image from an iPad camera can be emailed directly from the app. For more information go to the App Store.

ORGANIZATION OF THE SURFACE MICROBE HUNTER

The Surface Microbe Hunter is a classroom curriculum that uses the exploration of surface microbes to teach students STEM-based skills that they will apply to five activities.

The activities are set up for 10 student groups. Activity 1, and the follow up - Activity 2, requires a total of 10 paddles. Activity 3, Activity 4 and Activity 5 each require 10 paddles. Ten TSA/RB BioPaddles (Code 5552) are provided in the Surface Microbe Hunter. The paddles may be used for one class activity or they may be divided between multiple activities. Additional paddles are available separately. If students have not done the activities that precede the chosen activity, students should read the background material from the preceding activities. Suggested investigation order:

ACTIVITY 1 & 2	Contact sampling (1) and identification (2)
ACTIVITY 3	Stand alone; builds on Activity 1 and 2
ACTIVITY 4	Stand alone, advanced activity
ACTIVITY 5	Stand alone; builds on Activity 1 and 2

The Student Guide and Teacher Guide include background materials and instructions for each of the five activities. The Teachers Guide also includes recommended topics for student review and answers to student questions. Resources for the activities - including microbe identification guides, additional activities, and PowerPoint presentations on related topics– are provided on the Surface Microbe Hunter CD. Digital files can be viewed directly by the students or files can be printed and distributed.

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TEACHER NOTES and answers to student questions are printed in **BLUE**.

Active links to external web sites and files on the CD are printed in **ORANGE**.

The BioPaddles® Colony ID™ Lite App and BioPaddles® Colony ID™ App aid students in the presumptive identification of microbe growth on BioPaddles® with a large library of microbe images, ID guides, and resource materials. A report containing a full color image from an iPad camera can be emailed directly from the BioPaddles® Colony ID™ App. For more information go to the App Store.

NOTE: Example surface paddle growth images can be printed or uploaded (through iPhoto) to the BioPaddle™ Colony ID™ app for comparison to known standards.

CD CONTENTS

The Surface Microbe Hunter CD contains:

STUDY GUIDES (PDF)

Surface Microbe Teacher Guide
Surface Microbe Student Guide

BACKGROUND INFORMATION (PDF)

About The TSA/RB BioPaddle
BioPaddles Sampling Procedures
BioPaddle Storage and Disposal
Cleaners, Sanitizers, and Disinfectants
Colony Characteristics
Enumeration
Estimating Colony Size
Microbial Diversity
Microbe Motility
Preparing Dilute Solutions
Reproduction and Colony Formation
TSA/RB BioPaddle Tech Document

SURFACE MICROBES IDENTIFICATION GUIDE (PDF)

SURFACE MICROBE PRESENTATION (PowerPoint)

STEM ACTIVITIES

Do Antimicrobials Keep Products Cleaner?
How Clean Are Kitchen Sponges
Are Spore-Forming Microbes Affected by Disinfectants?
Build an Incubator

SURFACE MICROBE HUNTER GLOSSARY

SURFACE MICROBE HUNTER PADDLE IMAGES

SURFACE MICROBE HUNTER PUSH PLATE PADDLE IMAGES

SAFETY

Students should follow these safety and handling guidelines:

- Wash hands before and after handling BioPaddles® and samples.
- Do not open incubated BioPaddle vials.
- Dispose of BioPaddles® properly.
- Once the BioPaddle is removed from the protective vial, do not allow it to touch anything other than the sample.
- Keep the paddle in the protective vial until it will be used and replace it as soon as possible.
- Wash hands before and after performing experiments or handling samples and biological materials.
- Wear appropriate personal protective equipment.
- Know where emergency equipment is and how it works.
- Understand the hazards and risks regarding working with living organisms.

Care should be taken when culturing microbes in the classroom. Pathogenic microbes may be found in school environments. Pure cultures can be obtained from biological suppliers. Experiments are structured so that the materials, temperature, and media that are chosen do not encourage the growth of the more undesirable microbes. However, proper techniques and protocols for working with microbes should always be followed and PPE should be worn when working with any microbes.

SURFACE MICROBES OVERVIEW

Topics Covered:

- Surface Microbes
- Surface Characteristics
- Microbe Food
- Microbial Diversity

ACTIVITY: ARE ALL SURFACES ALIKE? Introductory Level Investigation

MATERIALS NEEDED

OBJECTIVES

PREPARATION

PROCEDURE

1. REVIEW SAFETY AND HANDLING INFORMATION
2. REVIEW BACKGROUND MATERIALS
3. THE TSA/RB BIOPADDLE
4. DISCUSS THE BIOPADDLE TECHNIQUE FOR SAMPLING SURFACES
5. THE LABORATORY NOTEBOOK
6. DESIGN THE EXPERIMENT

Four surface types will be sampled by student groups. The students can choose the sample, the teacher can tell the students where the sample can be found or provide the sample.

Sample Types are:

- | | |
|-------------|--|
| 1) POROUS | marked with pores, fissures, or cavities which allow liquids or solids to accumulate (Examples: cork, rough-sawn lumber, sponge, rocks, concrete, concrete blocks, sand and soils) |
| 2) TEXTURED | uneven and repetitive (Examples: corduroy fabric, canvas, suede, plant leaves) |
| 3) ROUGH | marked with irregularities, projections (Examples: terry cloth washcloth, deep pile carpet, tree bark) |
| 4) SMOOTH | free from irregularities, roughness or projections (Examples: glass, laminate countertops, enamel, bathroom tile, plastic wrap, metal, paper, plastic, chemically-modified plastics, painted surfaces) |



7. PROCEDURE

1. Use a permanent marker to label the BioPaddle vial. Include:
 - Group name
 - Date
 - Surface type
 - Incubation location/temperature
 - Incubation time
2. Remove the paddle from the plastic vial by pulling gently while twisting clockwise. Hold the paddle by the handle. Do not touch the agar surfaces. Do not put the paddle down on any surface. Avoid touching the BioPaddle to any other object.
3. Touch the **Tryptic Soy Agar (TSA) (yellow)** side of the BioPaddle to the test surface. The paddle must contact the surface firmly for at least 15 seconds.
Touch the **Rose Bengal Agar (RB) (pink)** side of the BioPaddle to the test surface. The paddle must contact the surface firmly for at least 15 seconds.
4. Replace the paddle in the vial. Be sure the paddle is secured tightly in the vial.

Vials may be taped shut to remind students not to remove the paddles from the vial during incubation, observation, and analysis.

Students should wash their hands immediately after sampling.

8. INCUBATION

9. ANALYSIS

1. Remove the BioPaddle vial from the incubation location. Do not open the BioPaddle vial.
2. Use a hand lens to examine the agar surfaces. Are there any colonies present?
3. Create side and top perspective drawings of each agar surface in a laboratory notebook.

The growth on the BioPaddle surfaces can be photographed with a digital camera or an iPad to document colony growth.

NOTE: Rotating the BioPaddle while it is INSIDE the outer vial will remove any condensation and allow students an opportunity to view (and photograph) colonies without removing the BioPaddle.

4. Create a data table in their laboratory notebook.

Surface Type	_____
Colony Count (Mold)	_____
Colony Count (Bacteria + Yeast)	_____
Total Colony Colonies	_____
Number of Species	_____
Microbial Diversity Value	_____

5. Count the total number of mold colonies (fuzzy colonies) on the TSA agar. Count the total number of bacteria and yeast colonies (non-fuzzy colonies) on the TSA agar. Do not open the BioPaddle vial.

NOTE: Sometimes bacteria colonies are crowded closely together and individual colonies cannot be distinguished. This is called confluent growth. If that happens, assign an estimated count number. Assume that a

square centimeter of confluent growth would have a count of 50 colonies. The BioPaddle agar pad is 10 cm X 10 cm.

Use **Enumeration** as a reference. Use the grid feature in the **BioPaddles® Colony ID™ App** to count the colonies on the captured image of the paddle growth.

6. Record the total number of colonies for the TSA agar.
7. Count the number of species (kinds) are on the TSA agar.

NOTE: A detailed analysis of the microbes, which includes identification, is covered in Activity 2. To retard growth, store the inoculated and incubated BioPaddles at cooler temperatures (around 66°F/10°C) until use in Activity 2. Do not refrigerate.

Or if no additional analysis is to be performed:

Dispose of the incubated paddles. Twist to remove the paddle from the vial. Fill the vial to the 40 mL line with a 1:9 dilution of household bleach (5.25% sodium hypochlorite solution). Replace paddle in the vial. Allow 15 minute contact time. Remove the paddle. Discard bleach solution. Replace the paddle in the vial and discard.

8. Calculate the microbial diversity value. Use **Microbial Diversity** as a reference.
9. Share group data with other groups to get a composite of microbe diversity for all four surface types.

For Example

Microbial Diversity on Surface Types				
Surface Type	Student hypotheses will vary but should include the following:	Microbial Diversity Value	Number of Bacteria/ Yeast Colonies (CFU/10 cm ²)	Number of Mold Colonies (CFU/10 cm ²)
POROUS	1 <i>Klebsiella</i> spp. (BACTERIA) 2 <i>Pseudomonas</i> spp. (BACTERIA)	0.5	10,000	----
TEXTURED	1 <i>Enterococcus</i> spp. (BACTERIA) 2 <i>Trichosporon</i> spp. (YEAST) 3 <i>Mucor</i> spp. (MOLD) 4 <i>Fusarium</i> spp. (MOLD) 5 <i>Bacillus</i> spp. (BACTERIA)	2.5	500	25
SMOOTH	1 <i>Aspergillus fumigatus</i> (MOLD) 2 <i>Trichosporon</i> spp. (YEAST) 3 <i>fungal mycelia</i> (FUNGI)	1.5	50	4
ROUGH	1 <i>Pseudomonas</i> spp. (BACTERIA) 2 <i>Microoccus luteus</i> (BACTERIA) 3 <i>Enterobacter</i> spp. (BACTERIA) - confluent 4 <i>Xanthomonas</i> spp. (BACTERIA)	2.0	150	----

NOTE: One would expect the porous surface type to have a higher diversity value, but in this example, there are only two kinds of bacteria – few “kinds” but high numbers is not as diverse as many kinds and few numbers. Names of microbes are included as examples of what species could be expected – students will not presumptively identify colonies until Activity 2.

Student answers will vary but the following relationships should be pointed out:

- A microbe diversity value of 0.01 is VERY LOW.
- A microbe diversity value of 99.0 is VERY HIGH.

- Surfaces having an increased surface area promote higher microbe diversity.
- Surfaces with organic material have greater microbe numbers and higher microbe diversity than those that do not have organic matter.

10. WRITE AN ANALYSIS REPORT

GOING FURTHER

RESOURCES

Additional Activities follow:

- ACTIVITY 2** Identifying Surface Microbes
ACTIVITY 3 How Do Sanitizers Work On Surfaces?
ACTIVITY 4 Design A Sanitizing Protocol
ACTIVITY 5 Antimicrobial Surfaces



Activities 2-5

Students can use the **BioPaddles® Colony ID™ Lite App** or **BioPaddles® Colony ID™ App** for iPads to enumerate and indentify colonies, capture an image, and prepare and email a report.

The Free LaMotte BioPaddles® Colony ID™ Lite app lets students compare colony examples on BioPaddle agar types from 5 microhabitats (air, water, soil, surface and food). Also contains information regarding organisms, microbiological techniques, and more!

New **BioPaddles® Colony ID™** app has a library of over 250 images of 30+ microbes, ideal for presumptive identification. Images of microbial growth on BioPaddles® can be captured with the iPad camera and imported for a side-by-side comparison to the images in the reference library. Using the new Report function a report including a full color image can be prepared and distributed directly by email. Expanded resource materials include Fungi and Bacteria Microanatomy and Microbe Exclusionary Charts. Available for purchase through iTunes. Visit our web site at www.lamotte.com and click on BioPaddles for a direct link.