Part I – Monday Morning Crisis at Lincoln Elementary

Secretary: Principal Pinder! I think we have a problem!

Principal: Mrs. Jackson, what’s wrong? It’s Monday morning and too early in the school day for problems.

Secretary: I know it’s early but our fifth grade teacher, Ms. Hines is sick.

Principal: Sick? What’s happening?

Secretary: Well, Ms. Hines called in this morning saying that she was having severe stomach pain. She looked well when I saw her on Friday before the class left for their field trip. Ms. Hines said she felt a little tired on Sunday evening and then this morning she has a fever and stomach cramps.

Principal: Mrs. Jackson, that’s not unusual. Teachers get sick all of the time because of germs spread by kids coughing and not covering their mouths, and people not washing their hands.

Secretary: Yes, that is true. Nevertheless, we’ve had ten calls from fifth grader parents! Their kids are sick with the same symptoms as Ms. Hines.

Principal: Ten sick children? Oh, my. What’s happening?

Secretary: There goes the phone again, I need to go answer it!

Principal: Mrs. Jackson… call Ms. Hines. I know she is sick but I need to find out what’s happening.

Secretary: Will do!

After calling Ms. Hines, Mrs. Jackson returns to Principal Pinder’s office a few minutes later.

Secretary: Principal Pinder, Ms. Hines says she just returned home from the doctor’s office. The doctor has sent urine and fecal samples to the lab for analysis. She should have her results back by tomorrow afternoon. Until then, Ms. Hines is on bed rest until the diagnosis is confirmed and her symptoms subside.

Questions
1. What is happening to the elementary school class?
2. What are the symptoms described in the case?
Part II – Ms. Hines Calls Principal Pinder

Ms. Hines called the school and asked to speak to Principal Pinder.

**Principal:** Hello Ms. Hines. I am so sorry to hear you are sick. The phone at school has been ringing off the hook with parents letting us know their children are sick too.

**Ms. Hines (sounding very ill):** Principal Pinder … I feel horrible. On Friday, Mrs. Garner and I took our classes on a field trip to the Science Center and the park. My class had a wonderful time. We watched a few of the science demonstrations and then went outside to the park and the ponds. We played on the playground. A small group of my students went to the new fishing dock and others fed the ducks. Some of the kids even had a chance to ride the paddleboats and splash in the water. We reunited with Mrs. Garner’s class in time to eat lunch and then we all returned to school.

I must admit, I felt progressively tired this weekend. I thought it might be due to all of the extra work I did to organize for the field trip. I tried to rest on Sunday to be ready for school on Monday. This morning, however, I was awakened with stomach cramps. I now have a fever and diarrhea. The doctor said my symptoms are consistent with gastroenteritis, which he suspects is being caused by an *Escherichia coli* (*E. coli*) bacterial infection. I’m not sure how I could have been exposed to *E. coli*. He asked me questions about where I had been or if I had recently eaten or drunk anything that may have been undercooked or unpasteurized. He also asked if I had visited a day care center, nursing home, or recently traveled out of the country. He said that food or water contaminated with *E. coli* bacteria can cause my symptoms.

**Principal:** Well Ms. Hines, we’ve had ten parents call in already this morning. The parents have indicated a variety of symptoms which include their children feeling lightheaded, chills, fever, diarrhea, and being tired. Did both of the fifth grade classes go to the park on Friday?

**Ms. Hines:** Yes, there were 17 students in my class and 20 in Mrs. Garner’s class on the school trip.

**Principal:** Hmm. That means we may be getting even more calls. Thank you Ms. Hines. I hope you feel better soon!

**Questions**

1. What does the doctor think may be the cause of Ms. Hines’ illness?
2. There are many conditions with symptoms similar to those described in the story. Research the symptoms. What bacteria or pathogens, other than *E. coli*, might be responsible for these illnesses? What are some examples of other illnesses that can have similar symptoms?
3. How could a class that visited a park potentially be exposed to *E. coli* bacteria?
4. What are potential modes for bacteria transmission?
Part III – Is Everyone Sick?

Principal Pinder next called Mrs. Garner, one of the other fifth grade teachers. She was in her classroom getting ready to start teaching her class for the day.

Principal: Hello Mrs. Garner, this is Principal Pinder. Are any of your kids sick today?

Mrs. Garner: Hello Principal Pinder. No, no one is sick. All 20 of my students are present and as lively as ever. They are still talking about how much fun they had during last week's field trip.

Principal: Oh good, I was wondering because Ms. Hines and ten of her students are very ill. I'm curious, did her class do anything different?

Mrs. Garner: Well, both Ms. Hines' class and my class watched several science presentations at the Science Center. After the demonstrations, my class voted to go to the petting zoo and Ms. Hines' class wanted to go to the park beside the Science Center. We all met back at the picnic area in time for lunch. Other than that, we all were together for the rest of the day.

Questions

1. Why is there concern about other kids getting sick?
2. Why do you think Mrs. Garner's students did not become ill while Ms. Hines' kids did?
3. Mrs. Garner's class visited the petting zoo, could they also have become sick from microorganisms?
4. Based on the circumstances, do you think the Principal should isolate the students that went on the trip from other students at school until further information is gathered?
Part IV – The Investigation Continues

Principal Pinder quickly went to her computer and searched for pictures of the park and pond areas. Using Google Earth, she found the following photos of the four locations visited by the children. Figure 1 shows the playground area and duck feeding area. Figure 2 shows the boat dock and Figure 3 shows the fishing dock.

*Figure 1. The playground area and duck area*

*Figure 2. The paddle boat dock*
Principal Pinder then wondered if *E. coli* was at the park where Ms. Hines and her class played. She called a physician friend to find out more about fecal coliform and *E. coli*.

**Principal:** Dr. Skinner, I need your help. I have several students and a teacher who went to our local park on Friday. They are now ill with stomach cramps and fevers. Based on what I have learned so far, the class went to several areas in the park and then had lunch. How could they have gotten sick if they simply went to a public park?

**Doctor:** I’m sorry to hear about your students and your teacher. I think there is a simple explanation but the harder question may be figuring out the source of the bacteria that resulted in their illness. Water quality standards are established by the U.S. Environmental Protection Agency (US EPA) to protect humans from exposure to chemicals and biological agents that can cause illness. Microorganisms are always present in lake water, but they are usually harmless. Sometimes after excessive rain, we find that harmful microorganisms like Cryptosporidium, Giardia, viruses, and total coliforms, which include bacteria such as *E. coli*, can enter waterways. These bacteria can originate from human or animal waste. These microorganisms can cause an illness called gastroenteritis, or a GI infection. Symptoms of a GI infection include diarrhea, vomiting, and cramps. Disease-causing microorganisms in lakes can originate from the guts of humans or animals. When their untreated fecal material makes its way into recreational waters, bad things happen.

**Principal:** Like the situation I now have with my teacher and students?

**Doctor:** Exactly. To see if waters are polluted with fecal material, we measure coliform bacteria as a primary indicator. If waters lack coliforms, it’s OK to go boating, fishing, and swimming in rivers or lakes. Coliform organisms are found in the intestinal tract of people and animals and are usually present in fecal wastes at much higher concentrations than disease-causing microorganisms. The presence of coliform bacteria in a water sample indicates that the water may be contaminated with fecal material and probably contains other disease causing organisms.

**Principal:** So, if coliform, or specifically *E. coli* bacteria, are present in the water, and the water got on their hands

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*Figure 3.* The fishing dock.
when they splashed, and they then somehow ingested the bacteria, is that how they potentially could have become ill?

Doctor: Very likely. You mentioned they ate lunch? That’s a possible route of ingestion. I think your next step should be to figure out which area of the park your sick students and teacher visited. Then, confirm if they washed their hands before they ate lunch. You may also want to have water samples from the park analyzed to track down the bacterial source.

Questions

1. What is the US EPA?
2. What are examples of bacteria and pathogens of concern in water that can cause illness?
3. What are the sources for coliform bacteria?
4. If the water is contaminated, how could it have infected the students?
5. Do you think that if someone did not wash their hands the contamination would spread? If so, how?
6. What are the next steps for Principal Pinder to prevent further outbreaks in the future?
Part V – Finding the Source (Laboratory Activity)

Principal Pinder calls your environmental consulting firm, Envirosafe Water Testing Services, for help with the water analysis. There are several methods that can be utilized.

**Positive/Negative Test**

Positive/negative tests are used to ascertain if a sample contains the bacteria you are investigating (positive) or the absence of the bacteria you are investigating (negative).

![Figure 4. Positive/negative test. Glowing indicates positive for coliform.](image)

**Membrane Filtration**

Membrane filtration consists of filtering the water sample through a 47-mm filter and growing the bacteria for 24 hours in an incubator. Petri dishes containing liquid media or agar are used to culture the bacteria. If fecal coliforms, *E. coli*, or enterococci are present in the water samples, colonies will grow (see Figure 5).

![Figure 5. Examples of colonies grown in petri dishes using liquid media.](image)

For membrane filtration, a simple single filtration unit can be used (Figure 6). M-Endo broth is a media that will support the growth of total coliform bacteria and M-ColiBlue-24 broth is another liquid culture media that will culture both total coliform colonies (blue) and *E. coli* colonies (red). To use, water is first filtered through the filter paper using the filtration apparatus. Using forceps, the filter paper is transferred into the petri dish and the cultures are allowed to grow for 24 hours in an incubator at either 35° C or 41° C based on the medium type.
Agar media instead of liquid media can be purchased from several suppliers to use with the petri dishes. Figure 7 shows *E. coli* and enterococci bacteria cultured on agar plates. *E. coli* colonies are purple and Enterococcus colonies are blue with pink halos.

**IDEXX Colilert and Enterolert Assays**

IDEXX Colilert and Enterolert assays are a liquid based method where Colilert powdered medium is used for total coliform and *E. coli* while Enterolert media is used for enterococci. These methods are more expensive than membrane filtration. To use, powdered media is added to 100-ml of the sample and shaken until dissolved. Samples are then poured into the IDEXX trays and heat sealed using the Quanti-Tray sealer. After sealing, the trays are placed in the incubator for 24-hours. The Colilert trays will turn yellow in natural light if total coliform bacteria are present and will fluoresce under black-light if *E. coli* is present. The Enterolert medium follows the same method as Colilert and the trays will fluoresce in black-light if positive for enterococci. The number of yellow wells or glowing wells are counted to determine the bacterial concentrations in the water sample. Figure 8 shows a picture of the Quanti-Tray sealer and two Quanti-Trays that are positive of total coliform and *E. coli*.

The US EPA sets the guidelines for the levels of total coliforms, enterococci, and *E. coli* bacteria that can be in lakes, rivers, or ponds used for recreational uses. Recreational water quality must meet certain criteria for safe use as presented in the table below.

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Figure 6. Apparatus for membrane filtration method

Figure 7. Agar plates with *E. coli* (purple) colonies and enterococci (blue) colonies.
**Criteria Elements**

<table>
<thead>
<tr>
<th>Indicator</th>
<th>Estimated Illness Rate (NGI): 36 per 1,000 primary contact recreators</th>
<th>OR</th>
<th>Estimated Illness Rate (NGI): 32 per 1,000 primary contact recreators</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enterococci—marine and fresh</td>
<td>35</td>
<td></td>
<td>30</td>
</tr>
<tr>
<td>E. coli—fresh</td>
<td>126</td>
<td></td>
<td>100</td>
</tr>
</tbody>
</table>

Duration and Frequency: The water body GM should not be greater than the selected GM magnitude in any 30-day interval.

**Source:** US EPA Recreational Water Quality Criteria (2012)

Values provide the geometric mean (GM) for the number of bacteria colonies, or colony forming units (CFU) per 100 ml of water. The geometric mean is the mean value for the samples. The NGI represents the number of people who develop a gastrointestinal illness (GI) infection per 1000 people using the recreational water. States may choose to use the 36 people per 1000 recreator standard or the 32 people per 1000 recreator standard to determine the acceptable levels of bacteria for a body of recreational water. A good rule of thumb to use is 200 CFU/100 ml E. coli or 33 CFU/100 ml enterococci for safe recreational water use by recreators. In terms of the amount of E. coli and enterococci in the recreational water, the chart is used to provide guidance for bacteria levels.

**Laboratory Activity**

Your consulting firm is going to help Principal Pinder by analyzing water samples from the pond to determine which location is the source for the fifth grade class illness. Review the areas identified in Part III of the case study. You will receive samples from each of the locations visited by the sick students and their teacher.

**Questions**

1. Review the three methods for assessing water samples. Discuss the ease of use and the tools needed for the analysis.
2. Which tests could be performed by a novice?
3. Which would require higher levels of technical knowledge?
4. Research the methods to determine the EPA approved methods for bacteria analysis.
5. Of the three methods, which do you feel would be the most accurate and informative?
6. What are the standards required by the US EPA for safe recreational use of water?