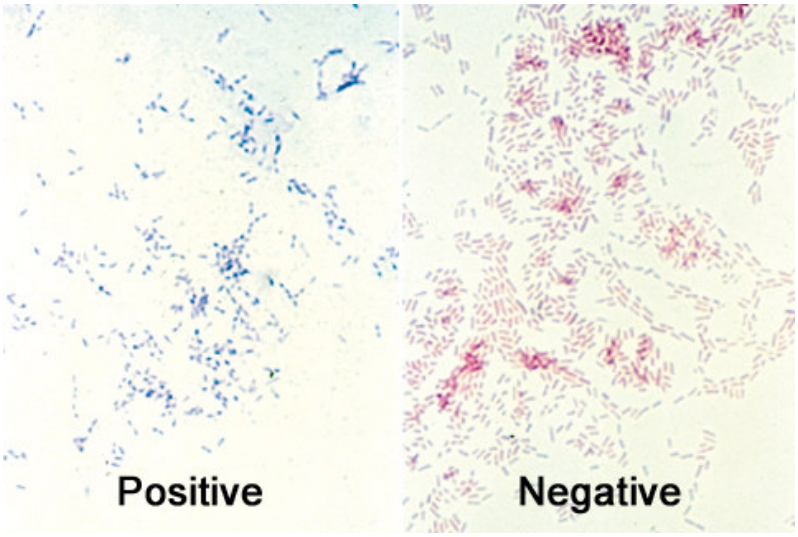
Clue # 1:

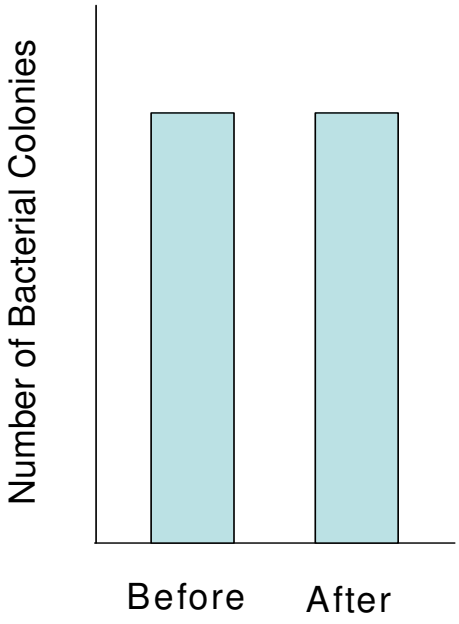
Bacteria can be divided into two groups (gram positive and gram negative) based on their appearance after iodine staining. This method was developed in 1884 (OVER 100 YEARS AGO) but is still commonly used to distinguish among different types of bacteria. Below is a key to the staining:



You make a slide of the bacteria and soak it in iodine. The bacteria that you are investigating turn **pink**.

Next you run a quick test to determine the shape of the bacteria. You add an antibiotic to the bacteria which **kills only round-shaped bacteria.**

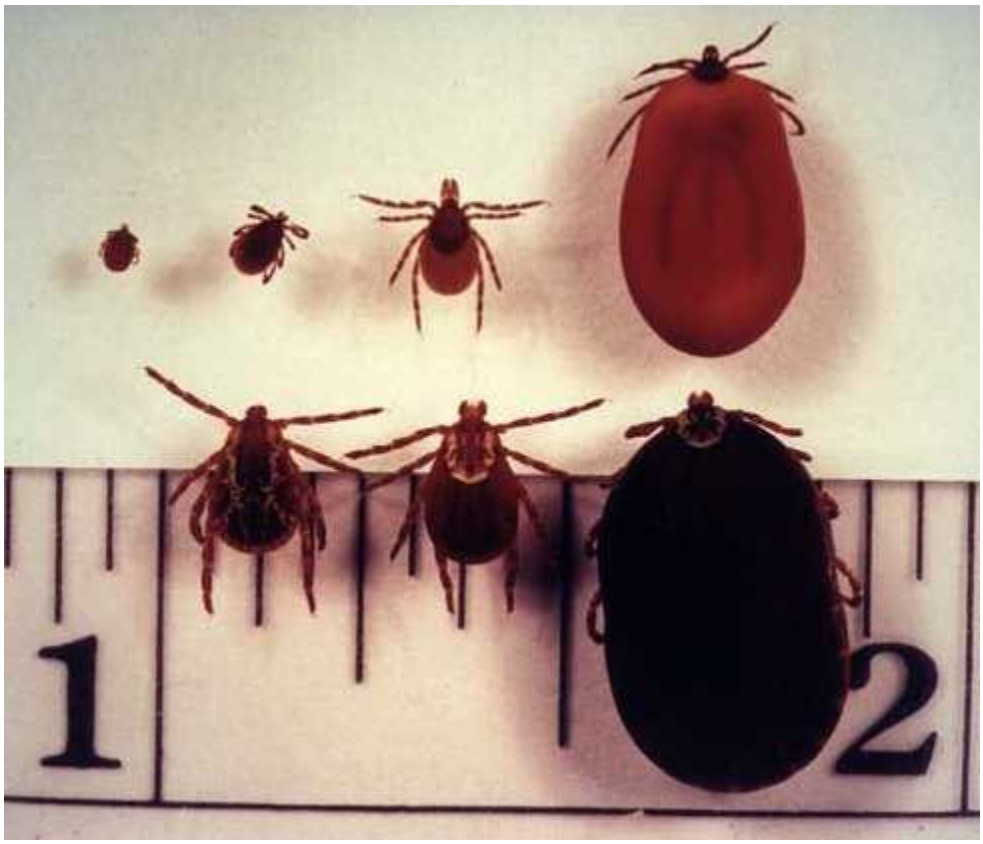
Here are your results before and after the addition of the antibiotic:



Clue # 2: You obtain the doctor's records for two of the victims of this disease and look for similarities.

Patient # 1: Victim is a 55 year old female nurse from Arizona with no history of health problems. The autosopy results showed a slight skin rash but no other symptoms. Patient had an extremely high fever when she arrived at the hospital.

Patient # 2: Victim is a 32 year old male gardener from Colorado with a history of smoking. Victim had been sick for 5 days with a high fever. Autopsy revealed a large reddish tick embedded in the victim's skin, less than ¼ inch wide. You notice a small rash around the bite that indicates the tick had been feeding for over 24 hours. You pull out your tick identification chart to see whether it's a deer tick or dog tick.



Top Row: *Ixodes scapularis (dammini*), the deer tick. Left to right: nymph, adult male, adult female, engorged adult female (a female which as just fed).

Bottom Row: *Dermacentor variabilis*, the American dog tick. Left to right: adult male, adult female, engorged adult female. Adults are most common in May, June, and July.

Clue # 3: You visit the victim's homes to interview their families about any animals they may have contacted recently. First you speak to Victim # 1's brother:

You: *I'm so sorry about your sister's passing. I know it must be hard to answer these questions right now, but you are doing an important service for other people who might be exposed to this deadly disease. Did your sister have any pets?*

Brother: *No, she liked to feed and watch birds, but she usually just ended up feeding squirrels instead. She didn't mind them though.*

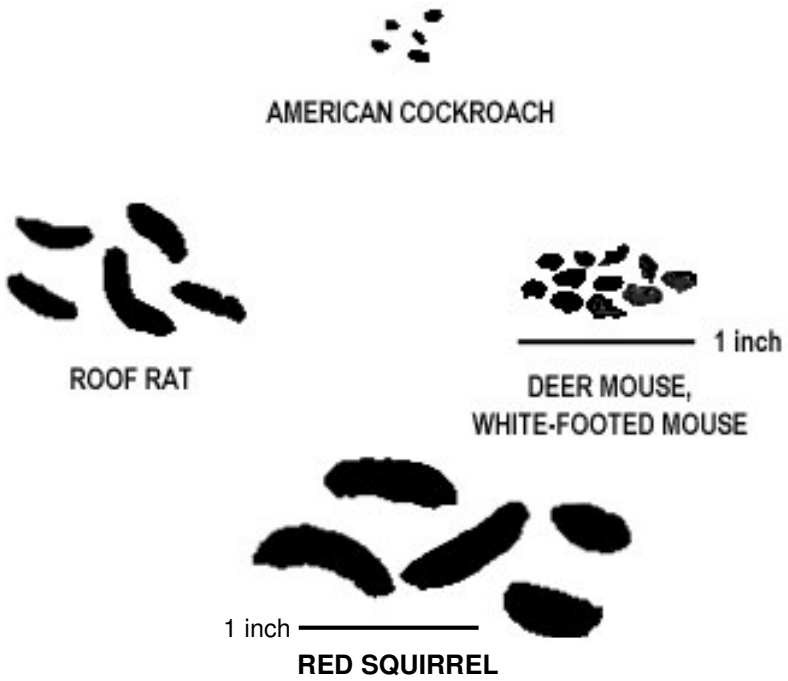
You: *Would she have come into contact with any other animals?*

Brother: *Let's see. She took care of one of her neighbor's dog a few weeks. That's about all I can think of.*

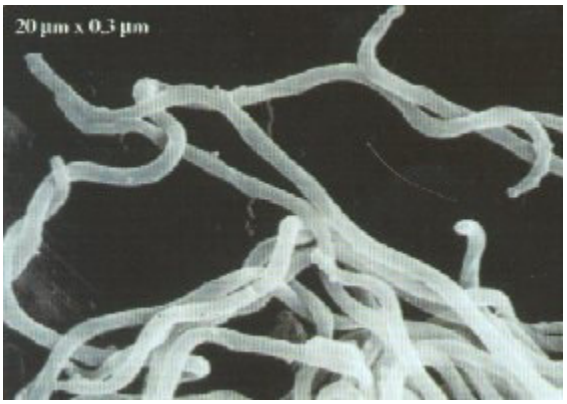
You: *What about at work? Would she have contacted any animals there?*

Brother: *I know she just took care of a wildlife rehabilitator who came in sick. I wonder if she picked up something from him.*

Next you head to Victim's #2 workplace to speak to a co-worker. The Gardening Agency appears to be closed for the day, so you leave feeling frustrated. As you turn to go, you notice a shed where the gardening tools are stored. You find the shed unlocked. You search inside and find it covered with animal scat (poop). You pull out your ruler and measure the pieces. Each piece is almost 1 inch long.



Clue # 4: You take out your electron microscope to get a final result on the shape of the bacteria. You take a look at a sample from a patient and see the following. You can now determine the true shape of the bacteria.



You look up which diseases deer ticks commonly carry and find the following from your research. You examine the information carefully to see if you can connect it to any of the earlier clues:

Rocky Mountain Spotted Fever

RMSF is a disease caused by rickettsia bacteria. A tick needs to be attached for four to six hours in order to transmit RMSF to its human host. The first symptoms noticed are usually severe headache, chills, fever, muscle aches, nausea, vomiting, and other flu-like symptoms. These first symptoms usually start 2 to 12 d after the tick bite. By the third day after the bite, a red rash develops on the wrists and ankles, in most cases, and often spreads to the entire hand or foot. A blood test is needed to confirm the disease.

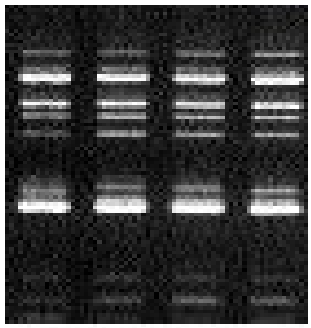
Lyme Disease

Lyme disease is caused by the spirochete bacteria Borrelia burgdorferi. A tick must be attached for at least 24 hours in order to transmit the disease organism to the host. The disease initially develops as an oblong rash, usually 2 or more inches in size, with a clear center that develops at the site of the tick bite; however, only 70% of people who are bitten by the tick develop this symptom. Within two days to a few weeks later people usually develop flu-like symptoms such as nausea, headache, and fever.

Clue # 5

Bacteria have “fingerprints” contained in their DNA. Each bacteria has a unique pattern of lines that form when you chop up its DNA and visualize it. To confirm the suspected culprit, you perform a DNA analysis samples collected from the four victims’ throats. You get the following results:

Bacterial DNA found in four victims:



1 2 3 4

You then compare these patterns to DNA fingerprints from three known bacteria that cause similar types of disease.

Rickettsia rickettsi

Francisella tularensis

Borrelia burgdorferi

Clue # 6

Press release, National Weather Service:

Rainfall in Colorado and Arizona were unusually high this year in the areas where all four victims are from.

Location	Year	Rainfall in Inches
Denver, Colorado	2001	40.7
Denver, Colorado	2002	38.3
Denver, Colorado	2003	**62.4**
Denver, Colorado	2004	41.6
Falstaff, Arizona	2001	28.4
Falstaff, Arizona	2002	31.6
Falstaff, Arizona	2003	**60.1**
Falstaff, Arizona	2004	39.4

You begin to think about how this might influence the squirrel population, and you pull out the following graph from a recent scientific article.