Experiment 3: Bacterial Behavior- Motility and Chemotaxis

One of the distinguishing characteristics of animals is their ability to move in response to stimuli that originate from within their own bodies or from the outside world. Many of us are attracted to the smell of fresh-baked chocolate chip cookies and repelled by the aroma of a recently antagonized skunk. An animal moving rapidly toward an object it recognizes as food or fleeing a harmful chemical are examples from the repertoire of responses known as animal behavior. Even the simplest and smallest animals exhibit behaviors within their own sensory capabilities.

But what about microbes? Microbes don't have what we typically associate with behavior, such as eyes, ears, noses, arms, legs, let alone a nervous system to interpret sensory signals. We spend a lot of time in the lab looking at colonies on hard agar surfaces. The bacteria are seemingly stationary and remain within the boundaries of the colony. You rarely see bacteria climbing over the side of the petri dish and onto the bench (although at least one species of bacteria, *Proteus mirabilis*, can do just that). Other than *Proteus*, most bacteria appear to be sedentary. However, this picture is misleading for many bacterial species. Not only can bacteria move under their own power, but they can also decide what to move towards and what to avoid. The *Proteus* that crawled over the side of their petri dish were seeking light.

Bacteria have several means of self-propelled locomotion: swimming, swarming, gliding, and twitching. The most common form of bacterial motility is powered by whip-like appendages called flagella that extend from the surface of the bacterium. Bacterial flagella are made completely of proteins. They are anchored in the cytoplasmic membrane by a protein assembly that works much like a rotary motor. In *E. coli*, the motor powers rotation of the long flagellum; counterclockwise rotation propels the bacterium forward, while clockwise rotation results in tumbling, non-directional behavior. Therefore, swimming *E. coli* move in a run and tumble fashion, moving in one direction, then tumbling and moving in a different direction.

For all motile microbes studied to date, the ability to move is coupled with the ability to move toward attractants and away from repellents. Bacteria use specialized membrane-bound sensory proteins to detect small changes in the concentration of chemicals in the environment. When the surrounding environment is homogenous, the bacteria move in random directions. However, when a localized attractant or repellent is present, the sensory receptors bind to the attractant or repellent initiating a signal transduction pathway that ultimately controls flagellar rotation. Attractants cause the flagella to rotate in the counterclockwise direction, prolonging the run phase of run-and-tumble. As a result, bacteria move toward an attractant in a biased random walk. Repellents cause the flagella to rotate in the clockwise direction, stopping motion and favoring tumbling, until the direction of the run moves the bacterium away from the repellent.

Bacteria respond to a diverse range of stimuli including nutrients, toxins, light, temperature, osmolarity, and even electrochemical potential. Directed motility, or "taxis", is used not only to find that perfect environment- enough food, not too hot, not too cold -

but also to allow bacteria to find niches in a host. *Helicobacter pylori*, the bacteria responsible for stomach ulcers, use chemotaxis to find the mucus lining of the stomach. Rhizobial species use chemotaxis to find roots of legumes with which they form a symbiotic relationship, getting nutrients from the host plant and providing nitrogen to the plant.

How do you know if a bacterium is motile? Bacterial flagella are too narrow to be seen in light microscopes like those we have for the lab class. However, flagella can be seen with electron microscopy or with special stains that coat the flagella making them visible. You can also determine if a bacterium is motile by observing its behavior. Does it zip around on your microscope slide, instead of flowing gently in the suspension? Motile bacteria appear to be moving with a purpose. You can also observe bacterial motility on semisolid agar in which the bacteria swim through holes in the agar. Because many bacteria move at once, areas that they occupy become cloudy. Non-motile bacteria remain in one place and don't spread across a plate of semisolid agar. If you have a motile bacterium, what chemicals, signals, and conditions attract or repel it? These are fundamental questions that we will explore in this lab module.

Resources

You can find basic information about flagella and chemotaxis in any microbiology textbook.
Online resource with an overview of bacterial motility, flagella, and taxis:
http://textbookofbacteriology.net/structure_2.html

Key concept

Bacteria behave in response to stimuli.

Challenge

We will use two different assays to determine if your pets are motile and chemotactic . We will also provide you with a plate of swimming *E. coli* and an assay to determine if a substance is an attractant or a repellent. **Develop a hypothesis about *E. coli*'s responses to different compounds- we will provide you with some sugars and amino acids. You are also welcome to bring in other substances that you would like to test.**

Key Questions

- What advantages does chemotaxis provide for a bacterium?
- High concentrations of repellents increase the tumbling time of *E. coli*. How does this help the bacteria move away from the repellent?
- Different bacteria chemotax towards different chemicals. Why might this be?
- Some bacteria are not motile. What factors in their environment might allow them to flourish without self-propelled motility?
- *Penicillium notatum* is not motile. However, it spreads over the surface of a petri dish and can even spread from one piece of bread to another. How might either of these behaviors occur in the absence of self-propelled motility?

References:
Adler, J. 2011 My life with nature. *Ann Rev Biochem* 80: 42-70.
Wadhams, G.H., Armitage, J.P. (2004) Making sense of it all: bacterial chemotaxis. *Nat Rev Mol Cell Biol* 5:1024-37.