Week 2: The Rate of Things

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1 Introduction

Last week we got a feel for the spatial scale of some biological organisms. This week, we are going to examine the temporal scale of some biological processes.

1.1 Goals for this week's lab:

The goal of this lab is to look at some of the most important biological processes. You will observe cell growth and division as well as the motion of some of their organelles. In order to look at these processes you will use the microscope but in order to keep track of the times involved, you will have to take images at regular time intervals. For this reason, make sure you familiarize yourselves better with the camera software that you will be using. In addition to the microscope, you will also learn how to plate cells at different concentrations and you will make optical density (OD) measurements as well.

The organisms and/or processes you will look at are briefly described in the following section.

2 What is Optical Density (OD)?

From Wikipedia, optical density (OD) is the absorbance of an optical element for a given wavelength λ per unit distance:

$$OD = \frac{A_\lambda}{l} = \frac{1}{l}\log_{10}(\frac{I_0}{I}) \tag{1}$$

where l is the distance the light travels through the sample (i.e. the sample thickness) measured in cm, A_λ is the absorbance at that wavelength, I_0 is the incident light beam intensity and I is the transmitted light beam intensity. Therefore, the lower the intensity of the transmitted beam, the higher the OD (the more concentrated the sample). So, one way of quantifying

how concentrated the sample is is by looking at the OD. In order to translate the OD measurement into a number of cells measurement we use the following formula:
10^8 cells /0.1OD

3 The Biological Organisms

Below we give a list and a brief description of the biological organisms we will observe this week.

***Escherichia coli* (*E. coli*):** One of the most studied prokaryotic organisms is *E. coli*. You can consider this as the unit of measurement to which all other prokaryotic organisms can be compared. It was discovered by Theodor Escherich, a German pediatrician and bacteriologist. *E. coli* is one of the main species of bacteria that live in the lower intestines of mammals, known as gut flora. Specimens have also been located on the edge of hot springs. According to US Department of Health and Human Services Centers for Disease Control and Prevention, the E. coli strain O157:H7, one of hundreds of strains of the bacterium E. coli, causes illness in humans. Presence in surface water is a common indicator of fecal contamination. It belongs among the Enterobacteriaceae, and is commonly used as a model organism for bacteria in general. One of the root words of the family's scientific name, "enteric", refers to the intestine, and is often used synonymously with "fecal" (Wikipedia).

For this part of the lab, you will look at the growth of *E. coli*cells. You might want to take snapshots at regular time intervals in order to observe the growth. In the end, what you will want to obtain is a graph of the number of cells in the microscope's field of view vs. time.

***Saccharomyces cerevisiae* (*S. cerevisiae*):** *S. cerevisiae* is a species of budding yeast. It is perhaps the most important yeast owing to its use since ancient times in baking and brewing. It is believed that it was originally isolated from the skins of grapes (one can see the yeast as a component of the thin white film on the skins of some dark-colored fruits such as plums; it exists among the waxes of the cuticle). It is one of the most intensively studied eukaryotic model organisms in molecular and cell biology, much like *Escherichia coli* as the model prokaryote. It is the microorganism behind the most common type of fermentation. It reproduces by a division process known as budding. It is useful in studying the cell cycle because it is easy to culture, but, as a eukaryote, it shares the complex internal cell structure of plants and animals. *S. cerevisiae* was the first eukaryotic genome that was completely sequenced. The genome is composed of about 13,000,000 base pairs and 6,275 genes, although only about 5,800 of these are believed to be true functional genes. It is estimated that yeast shares about 23% of its genome with that of humans. "Saccharomyces" derives from Greek, and means "sugar mold". "Cerevisiae" comes from Latin, and means "of beer" (Wikipedia).

Again, for this part of the lab, take several snapshots of the yeast cells at periodic time intervals. You will also learn how to plate these cells at different concentrations and you

will also learn how to get the concentration by doing OD measurements. In the end, again, you will want to create a graph of the number of yeast cells vs. time.

***Dictyostelium*:** *Dictyostelium* is a cellular slime mould. When food (normally bacteria) is readily available the cells take the form of individual amoebae, which feed and divide normally. However, when the food supply is exhausted, they aggregate to form a multicellular assembly, called a pseudoplasmodium or slug (not to be confused with slug the animal). The slug has a definite anterior and posterior, responds to light and temperature gradients, and has the ability to migrate. Under the correct circumstances the slug matures forming a fruiting body with a stalk supporting one or more balls of spores. These spores are inactive cells protected by resistant cell walls, and become new amoebae once food is available (Wikipedia).

Here you will look at the life cycle of dicty. The individual cells will from fruiting bodies when they run out of food!

***Stentor polymorphus*:** Stentor protozoa are single-celled animals that grow to a length of 1.5 to 2 millimeters, much larger than many of their fellow multi-cellular aquatic organisms. Often, stentors will attach the lower portion of their pod to debris and assume a trumpet-like shape. The circle of tiny cilia surrounding the trumpet rim beat continuously and serve to create localized convection currents in the water to draw smaller organisms into the rim or mouth (cytostome). Stentors vary in color, depending upon their diet, but they can appear green, blue, or reddish-yellow.
(http://micro.magnet.fsu.edu/primer/techniques/hoffmangallery/stentor.html).

The trumpet-like shape is what gave this organism its name. Stentor was a figure in the Greek mythology. He was a herald of the Greek forces during the Trojan War, made famous by his loud voice (the adjective "stentorian" means loud-voiced). He died after he was defeated by Hermes in a shouting contest.

Here, try to take a video of the cilia motion located at the rim of the trumpet. This motion helps Stentor draw food particles down into the gullet. Try to get a feel for the period of this motion

***Danio rerio* (*D. rerio* or Zebrafish):** The zebrafish is a tropical fish and an important model organism for studies of vertebrate development and gene function. They are commonly used for these studies because the reproduce easily. Also, the embryo is transparent, which helps when studying the organ shape and function (Wikipedia).

For the case of the zebrafish, you can look at and study the cilia motion and the beating of its heart.