The Cell as a Machine

Lecture 1 Overview and Outline

Goals of Course: To provide a working understanding of the basic cell functions physical and chemical principles underlying them. In practical terms, we will attempt to solve a number of important problems relevant to replication, transcription, translation, translocation, motility, and other important functions.

Definition of a Cell: Our working definition of a biological cell is a self-contained unit that is capable of duplicating itself given the proper nutrients and environment. For the purpose of this course, we will focus on mammalian fibroblastic cells but not exclusively.

History of Cells: There is geological evidence of cellular life over the last 2 billion years on earth. Whether the first cells formed directly from some primordial soup or came from some other planet, is not of interest here. What is of concern is how current cell systems have reached their present state of sophistication. Darwinian evolution suggests that survival goes to the fittest and it is worthwhile for us to consider what this means in practical terms for cellular functions. The process of Darwinian selection is analogous to the process of evolution of automobiles. In the late nineteenth century a basic automobile was developed and since that time billions of automobiles have been produced with increasing sophistication. Refinements of the automobiles such as fuel injection, seatbelts and even cupholders are incorporated into most modern vehicles because people prefer to buy cars with those features. Cars without those features can work but there is an incremental preference for cars with those features.

Cell Optimization and Robustness: Because there have been countless numbers of organisms that have come and gone, nature has selected cellular systems that can survive and carry forward the genetic material that defines cell composition through many adverse conditions. The property of robustness is therefore strongly favored in cells. Robustness is defined as the ability to function properly under a variety of conditions, including changes in 1. number of proteins per cell, 2. salinity and pH, 3. temperature, 4. nutrient level, and 5. environmental factors.

Time to Extinction: It is useful to consider how long the genes coding for less competitive proteins will remain in the gene pool.

Steady State System: In a stable ecosystem, the population of any given species will assume some average level over cyclic fluctuations. For example, the mouse population on a hypothetical island is on average 100,000 and they have a generation time of six months. Assume that a mutation in a protein gives the mice a 1% survival advantage as a heterozygote and 2% as a homozygote over the original protein in one generation, which means for the sake of calculation that for every 100 mice in the first generation 102 homozygous mutants will reach the second generation. After 1000 generations and several cycles of population growth and contraction, the population of animals with homozygous original gene will be very small (less than 3% of the total and the number of homozygous animals will be even less (it should be pointed out that inbreeding of homozygous animals will increase the rate of decline)).

Feast-famine cycles: More common in history is the expansion of the population of a given species and then a dramatic decline in that population due to the expansion of a predator population, an environmental change, or a disease. In those cases the selection pressures can be much more severe and over 90% of a given population can be lost in a brief period. Those situations are difficult to model because they select for markedly different factors, e.g. the decimation of buffalo populations by early settlers and of Hawaiians by European diseases. The high selection pressures in these situations can often select for mutant genes in the population that would otherwise not be favorable, e.g. malaria selection of G6PD deficiency and sickle hemoglobin. In such cases, there may be clonal selection of individuals who have particular genes.

1. Number of Proteins per Cell. The number of molecules of any protein per cell is often very small (as low as tens of copies per cell), which means that large variations in the concentration can occur randomly (e.g. the noise in counting radioactive decay processes scale as the inverse of the square root of the number of events, thus for 100 counts the error is ±1/10 or ±10%). Further, as cells divide the partitioning of proteins between the two daughter cells is often uneven. If a function depends directly upon the number of proteins per cell, then the two daughter cells will differ in function. Where it has been studied in detail in the chemotaxis of bacteria, the concentration of many proteins can vary over a thousand-fold without altering the ability of the cells to chemotax ([Alon, 1999 #1]).

Calculations of Cell Size

Eukaryotic cells range in size from about 4 microns to millimeters (for larger syncytial cells). An average size for the common cells used in cell culture (HeLa or mouse 3T3 cells) is 15-20 microns in diameter for a suspended cell (volume $4/3\pi r^3$ = 4000 μm^3 or 4 x 10^{-9} cm^3). In contrast, bacteria (prokaryotic cells) such as Eschericia coli are cylinders of about two microns in length and 0.8 micron in diameter (volume $l\pi r^2$ = 1 μm^3 or $1x10^{-12}$ cm^3). The concentration of cytoplasmic proteins is about 180 mg/ml. If we assume that the average molecular weight of proteins is 50 kDa, then the overall concentration is 3.6 mM or 2 $x10^{18}$ molecules per ml (alternatively, 8 x 10^9 molecules per eukaryotic cell or 2 x 10^6 molecules per prokaryotic cell. If we carry this approximate calculation further and introduce the number of different protein molecules in a cell (10,000 for the eukaryotic and 2,000 for the prokaryotic), then the number of molecules of any given protein will be on the order of 10^5 and 10^3, respectively.

2. Salinity and pH. For bacteria and many fishes, the ability to withstand osmotic shock is critical as is the need to survive changes in ion content of the surrounding medium and pH. A sudden flood can decrease the salinity in a salt marsh by over two-fold and the organisms must adapt rapidly or lyse when water moves into the cells. We will discuss cell volume control and osmotic pressure as a regulator of ion channels. Also, there are many events that can cause cells to lyse, including osmotic pressure, mechanical tears and detergents. There is a large reservoir of internal lipid in most cells that can use to seal over a leak in the plasma membrane.

3. Temperature. Freeze-thaw cycles are common for organisms in Northern climates and cells must be able to survive or be lost. Further, at low temperature some filament systems such as microtubules will disassemble which will block mitosis. Excess heat poses a different set of problems. Heat denaturation of proteins results in an excess of non-functional protein and insoluble aggregates that the cell must clear. Heat shock protein expression is stimulated at high temperature and under conditions of stress to the organism.

4. Nutrient level. Fluctuations in nutrient level must be accompanied by changes in cell metabolic levels to preserve viability. Storage of nutrients for hard times is critical for survival. Fat storage and glycogen storage are obvious adaptations but homeostatic mechanisms are in place to enable transitions from carbohydrate to fat metabolism, from metabolizing nutrients in the gut to stored material, and ultimately in starvation to recycling components from its own cells.

5. Environmental Factors. This is a catchall category that lumps together other factors that are critical for organism survival. Parasites (viruses, bacteria or larger organisms) and other infectious agents all contribute to the function of complex organisms, often synergistically but sometimes they kill the host when things go awry. At a beneficial level, viruses and bacteria can carry genetic information from one organism to another.

Cell State Effects on Functions

We often respond differently to the same stimulus when we are in different contexts. In a similar way, any cell has a history and a current context that will influence its response to a given stimulus. Thus as we consider cellular functions, we will try to define the state of the cell but if no state is specified, you can assume that we are talking about HeLa cells in interphase.

Critical Aspects of Eukaryotic Animal Cell Functions

As a self-replicating machine, the cell must contain all of the functions needed to propagate its DNA. In the case of a HeLa or other mammalian cell in culture, the cell is in a relatively constant environment with sufficient soluble nutrients and space to grow. Thus, the cell has to grow and to maintain its current state (ion and water transport are needed to keep volume and ionic composition of the cell constant). Underlying all cellular functions is the fact that diffusive energy provides the basis for enzymatic activities as well as movements. We will first discuss the low Reynolds number of cells and diffusive processes. Because the propagation of the DNA is the most critical aspect of cell viability, we will then discuss DNA packaging in the nucleus, replication of DNA, and translation to RNA. A critical aspect of cellular function is the ability to compartmentalize regions of cytoplasm as well as maintaining a cell boundary, which often involves a membrane; therefore, we will discuss the important aspects of membrane function. The plasma membrane is structured by the cytoskeleton and the cytoskeleton is a critical component in organizing the cell.

Course Outline:

1. Introduction

2. How Nano-BioMachines Work (diffusion and transport) (Lectures 2 & 3)

3. DNA Packaging and Replication (Lectures 4, 5 and 6)

4. RNA Transcription and Processing (Lectures 7 and 8)

5. Lipid Bilayer and Plasma membrane (hydrophobic effect, Guoy-Chapman potential, mechanics, and diffusion in 2-D) (Lectures 9, 10, and 11)

6. Midterm (note that the course organization is different from last year)

7. Protein synthesis and processing (polymerases, processing (hybridization), ribosomes, and translocation) (Lectures 12, and 13)

8. Cytoskeleton, which defines a non-spherical shape and mechanical properties (polymer assembly, mechanics) (Lectures 14, 15, 16 and 17)

9. Glycoprotein and Secreted Protein Processing (Membrane transport, flow, and stabilization) (Lectures 18, and 19)

10. Endocytosis and Protein Degradation (Lecture 20)

11. Ion balance and volume regulation (membrane potential, osmotic pressure, and resealing) (Lecture 21)

12. Signal Integration (Biosystems Engineering) (Lecture 22)

13. Migration, Force generation and Chemotaxis (Lectures 23, 24 and 25)

14. Final Exam (Will contain problems from whole course with emphasis on last half)

Problem for First Lecture:
Calculate the number of generations to extinction of a gene for a native protein in mice that does not confer protection to a lethal disease in the presence of a mutant that will protect against the disease. Assume that 90% of the homozygous native protein animals die, whereas 60% of the heterozygous animals and 10% of the homozygous mutant animals die if an epidemic of the disease strikes. When the epidemic first strikes, the population is 10,000 with 20% of the mutant gene. Because the natural resources are not limiting, the animals that survive the disease will double in number each generation. The disease strikes once in each generation on average. (Remember that you can't have fractions of animals and if the calculations say that 7.9 animals will survive, then in reality only 7 do survive. Less that 1 means 0 or extinction). To give a hint, there are 400 homozygous mutant, 3200 heterozygous, and 6400 homozygous native animals before the first epidemic; and after the first epidemic, the number drops to 360 homozygous mutant, 1280 heterozygous, and 640 homozygous native animals.