MITOSIS AND MEIOSIS

Part 1. Observing Mitosis and Cytokinesis in Plant Cells

Materials
- prepared slide of onion root tip
- compound microscope

Introduction
The behavior of chromosomes during the cell cycle is similar in animal and plant cells. However, differences in cell division do exist. Plant cells have no centrioles, yet they have bundles of microtubules that converge toward the poles at the ends of a spindle. Cell walls in plant cells dictate differences in cytokinesis. In this exercise, you will observe dividing cells in the zone of cell division of a root tip.

Procedure

1. Examine a prepared slide of a longitudinal section through an onion root tip using low power on the compound microscope.
2. Locate the region most likely to have dividing cells, just behind the root cap.
   At the tip of the root is a root cap that protects the tender root tip as it grows through the soil. Just behind the root cap is the zone of cell division. Notice that rows of cells extend upward from this zone. As cells divide in the zone of cell division, the root tip is pushed farther into the soil. Cells produced by division begin to mature, elongating and differentiating into specialized cells, such as those that conduct water and nutrients throughout the plant.
3. Focus on the zone of cell division. Then switch to the intermediate power, focus, and switch to high power.
4. Survey the zone of cell division and locate stages of the cell cycle: interphase, prophase, prometaphase, metaphase, anaphase, telophase, and cytokinesis.
5. As you find a dividing cell, speculate about its stage of division, read the following descriptions given of each stage to verify that your guess is correct, and, if necessary, confirm your conclusion with the instructor.
6. Locate the nucleus, nucleolus, chromosome, chromatin, mitotic spindle, and cell plate when appropriate.

Part 2. Duration of mitotic stages:
If we assume that the entire cell cycle in an actively dividing region of a plant is about 24h, and if we further assume that the length of each stage is indicated by how many cells we observe in a particular stage, it is possible to determine the approximate length of time required to complete each stage of mitosis.

1. With the microscope set on a medium power, locate a region of the onion root tip squash with a number of dividing cells.
2. Count the total number of cells in the field of view and the number of nuclei in prophase, metaphase, anaphase, telophase. Record the numbers of each.
3. The total number of cells minus the total number of dividing cells gives the number of cells in interphase. Record this number.
4. The duration of each mitotic phase can now be estimated with the following equation:
   \[
   \text{Duration of stage} = \frac{\text{# of cells in stage}}{\text{total # of cells}} \times 24 \text{ hr} \times \frac{60 \text{ min}}{1 \text{ hr}}
   \]
The duration of a cell cycle varies tremendously from one cell type to another and between different types of organisms. The embryonic cells of any species are generally the most rapid dividing. A standard cell cycle generally has the following timing:

<table>
<thead>
<tr>
<th>Stage</th>
<th>Duration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prophase</td>
<td>20 min</td>
</tr>
<tr>
<td>Metaphase</td>
<td>40 min</td>
</tr>
<tr>
<td>Anaphase</td>
<td>5 min</td>
</tr>
<tr>
<td>Telophase</td>
<td>20 min</td>
</tr>
<tr>
<td>Interphase</td>
<td>22.5 h</td>
</tr>
</tbody>
</table>

How do your results compare with these?

**Part 3-Modeling Meiosis**

**Materials**
- 60 pop beads of one color
- 4 centrioles
- 60 pop beads of another color
- 8 magnetic centromeres

**Introduction**

Meiosis takes place in all organisms that reproduce sexually. In animals, meiosis occurs in special cells of the gonads; in plants, in special cells of the sporangia. Meiosis consists of two nuclear divisions, meiosis I and II, with an atypical interphase between the divisions during which cells do not grow and synthesis of DNA does not take place. This means that meiosis I and II result in four cells from each parent cell, each containing half the number of chromosomes, one from each homologous pair. Recall that cells with only one of each homologous pair of chromosomes are haploid (n) cells. The parent cells, with pairs of homologous chromosomes, are diploid (2n). The haploid cells become sperm (in males), eggs (in females), or spores (in plants). One advantage of meiosis in sexually reproducing organisms is that it prevents the chromosome number from doubling with every generation when fertilization occurs.

*What would be the consequences in successive generations of offspring if the chromosome number were not reduced during meiosis?*

**Interphase:** Working with group members, you will build a model of the nucleus of a cell in interphase before meiosis. Nuclear and chromosome activities are similar to those in mitosis. Discuss activities in the nucleus and chromosomes in each stage. Go through the exercise once together and then demonstrate the model to each other to reinforce your understanding. Compare activities in meiosis with those in mitosis as you build your model.

**Procedure**

1. Build the premeiotic interphase nucleus. Have two morphologically distinct pairs of chromosomes (2n = 4). Have one member of each pair of homologues be one color, the other a different color.
2. To represent G1 (gap 1), pile your four chromosomes in the center of your work area. The chromosomes are decondensed. *In G1, are chromosomes single-stranded or double-stranded?*
3. Duplicate the chromosomes to represent DNA duplication in the S (synthesis) phase. Recall that in living cells the centromeres remain single, but in your model you must use two magnets. *What color should the sister chromatids be for each pair?*
4. Duplicate the centriole pair.
Meiosis I

Meiosis consists of two consecutive nuclear divisions, called meiosis I and meiosis II. As the first division begins, the chromosomes coil and condense as in mitosis. Meiosis I is radically different from mitosis, however, and the differences immediately become apparent. In your modeling, as you detect the differences, make notes in the margin of your lab manual.

Procedure

1. Meiosis I begins with the chromosomes piled in the center of your work area.
2. Separate the two centriole pairs and move them to opposite poles of the nucleus.
3. Move each homologous chromosome to pair with its partner. You should have four strands together. How many tetrad complexes do you have in your cell which is $2n = 4$?
4. Represent the phenomenon of crossing over by detaching and exchanging identical segments of any two nonsister chromatids in a tetrad. Crossing over takes place between nonsister chromatids in the tetrad. In this process a segment from one chromatid will break and exchange with the exact same segment on a nonsister chromatid in the tetrad. The crossover site forms a chiasma (plural, chiasmata).
5. Move your tetrads to the equator, midway between the two poles. Late in prophase I, tetrads move to the equator.
6. To represent metaphase I, leave the tetrads lying at the equator. During this phase, tetrads lie on the equatorial plane. Centromeres do not split as they do in mitosis.
7. To represent anaphase I, separate each double-stranded chromosome from its homologue and move one homologue toward each pole. (In our model, the two magnets in sister chromatids represent one centromere holding together the two sister chromatids of the chromosome.) How does the structure of chromosomes in anaphase I differ from anaphase in mitosis?
8. To represent telophase I, place the chromosomes at the poles. You should have one long and one short chromosome at each pole, representing a homologue from each pair. Two nuclei now form, followed by cytokinesis. How many chromosomes are in each nucleus? Would you describe the new nuclei as being diploid (2n) or haploid (n)?
9. To represent meiotic interphase, leave the chromosomes in the two piles formed at the end of meiosis I.
10. Duplicate the centriole pairs. Meiosis II

The events that take place in meiosis II are similar to the events of mitosis. Meiosis I results in two nuclei with half the number of chromosomes as the parent cell, but the chromosomes are double-stranded (made of two chromatids), just as they are at the beginning of mitosis. The events in meiosis II must change double-stranded chromosomes into single-stranded chromosomes. As meiosis II begins, two new spindles begin to form, establishing the axes for the dispersal of chromosomes to each new nucleus.

Procedure

1. To represent prophase II, separate the centrioles and set up the axes of the two new spindles. Pile the chromosomes in the center of each spindle.
2. Align the chromosomes at the equator of their respective spindles. As the chromosomes reach the equator, prophase II ends and metaphase II begins.
3. Leave the chromosomes on the equator to represent metaphase II.
4. Pull the two magnets of each double-stranded chromosome apart.
   As metaphase II ends, the centromeres finally split and anaphase II begins.
5. Separate sister chromatids (now chromosomes) and move them to opposite poles.
   In anaphase II, single-stranded chromosomes move to the poles.
6. Pile the chromosomes at the poles.
   As telophase II begins, chromosomes arrive at the poles. Spindles break down. Nucleoli reappear. Nuclear envelopes form around each bunch of chromosomes as the chromosomes uncoil. Cytokinesis follows meiosis II.
   
   What is the total number of nuclei and cells now present?
   How many chromosomes are in each?
   How many cells were present when the entire process began?
   How many chromosomes were present per cell when the entire process began?
   How many of the cells formed by the meiotic division just modeled are genetically identical? (Assume that alternate forms of genes exist on homologues.)

This may seem like an insultingly simple exercise, but if you can demonstrate each of the various stages of mitosis and meiosis, you can be confident that you really understand the differences between the two types of cell division.

Web activities
http://biog-101-104.bio.cornell.edu/biog101_104/tutorials/cell_division.html