EDVOTEK®

AP BIOLOGY INVESTIGATION 7

EDVO-Kit: AP07

Cell Division: Mitosis and Meiosis

See Page 3 for storage instructions.

EXPERIMENT OBJECTIVE:

The objective of this experiment is for students to identify and differentiate various stages in mitosis and meiosis. Onion root tips are stained to identify the various stages and duration of mitosis. Students will also have an opportusnity to analyze mechanism involved with loss of cell cycle control in cancer. Meiosis and Crossing Over in *Sordaria* is also examined in this experiment.

Table of Contents

Experiment Components

Store the entire experiment at room temperature.

This experiment is designed for 10 lab groups.

Investigations I & IV
- 3-cm long Pipe cleaners (in 2 colors)
- Beads
- Small plastic bags

Investigations II
- Carbol-fuschin (Ziehl-Neelson) stain
- Lectin (phytohemagglutinin PHA-M from *Phaseolus vulgaris*)
- Microscope slides and covers
- 10 plastic cups (to grow onion root tips)
- Sand
- 50 ml conical tubes

Investigations III
- Karyotype pictures of normal individuals
- Karyotype pictures of patients 1, 2, and 3

Investigations V
- Pictures of *Sordaria fimicola* (Color pictures recommended)

Requirements

Investigations I & IV
- Colored pencils (2 colors)

Investigation II
- Microscope
- 10 green onions (or scallions) with roots
- Ethanol
- Glacial acetic acid (12 M)
- Hydrochloric acid
- Razor blades
- Scissors
- Scientific cleaning wipes (Kimwipes)
- Disposable gloves

Investigations III & V
- Photocopier

Background Information

MITOSIS AND MEIOSIS

The growth and development of every organism depends on the precise replication of the genetic material during each cell division. It is remarkable when one considers that as individuals we have all arisen from the fertilization of a single egg with a single sperm. From this single cell we develop into unique individuals with highly differentiated tissue types. The instructions for the precise timing of development, growth and maturation are all contained within DNA, which is organized as nucleotides encoding specific genes, which are organized into chromosomes. Each cell contains this set of information. Differential gene expression is what accounts for the obvious differences between the various tissue types that make up nerves, skin, muscle and organs such as the kidneys, liver and spleen.

The cell cycle, the sequence of events that encompass the period between the completion of one cell division until the end of the next division, involves both division of the cell's nucleus **(karyokinesis)** and division of the cytoplasm **(cytokinesis)**. There are two types of nuclear division: **mitosis** and **meiosis**. New body (somatic) cells are formed by mitosis. Each cell division produces two new daughter cells with the same number and kind of chromosomes as the parent cell. The formation of male and female gametes in animal cells or spores in plant cells is by meiosis. Gametes and spores will have half the chromosome number of the parent cells.

Interphase

Interphase, which begins when cell division ends and continues until the beginning of the next round of division, is organized into three phases. **G1**, is the first growth period of interphase. The nucleus and cell increase in size, and chromosomes are fully extended. The cell expends large amounts of energy in the synthesis of RNA and protein. During G1, the cell carries out normal functions specific to its type (i.e., nerve, liver, spleen). **S**, the next section of interphase, is marked by a dramatic rise in DNA synthesis, and synthesis of histones that are major cellular proteins bound to DNA. The cell is preparing for the beginning of mitosis. The chromosomes are becoming longitudinally doubled, with each chromosome consisting of two identical "chromatids." **G2**, the final segment, is marked by continued protein synthesis. A cell in interphase has a nucleus with one or more dark-stained **nucleoli** and a fine network of threads, the **chromatin**.

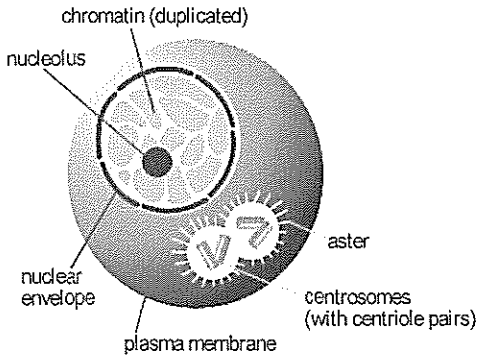


Figure 1 - Interphase

Background Information

Mitosis

Mitosis is the next phase of the cell cycle. It is the process of coordinated chromosome replication prior to cell division. It is essentially the same whether considering a simple plant or a highly evolved organism, such as a human being. The major function of mitosis is to accurately and precisely replicate genetic information, or chromosomes, so each daughter cell contains the same information. The enzymatic complex, a DNA polymerase, accomplishes this task with an average of less than one error, or one base pair change per 1×10^9, nucleotides synthesized. The human genome contains approximately 3.3×10^9 base pairs, so less than 3 errors would occur during a typical cell division.

The process of mitosis is an ongoing event that can be segmented into several identifiable stages. During the mitotic phase, a unique compliment of genes are activated. These genes encode proteins which act only transiently during mitosis and are absent from other phases of the cell cycle. In order, these stages are: **prophase**, **metaphase**, **anaphase**, and **telophase**. **Cytokinesis**, the actual process of cell division, occurs during telophase. In plants such as the onion, this is seen as the formation of the cell plate between the two **daughter cells**.

Prophase

In prophase, dramatic changes begin to occur within the nucleus of the cell. Chromosomes become thicker, shorter, and easily visible when stained under the light microscope. Two "**sister chromatids**" join near their middle at a structure called the **centromere**. The nucleolus, the site of active rRNA synthesis, and the nuclear membrane disappears. The mitotic apparatus, the **spindle**, begins to organize within the cell. **Microtubules** are slender rods of protein responsible for pulling replicated chromosomes towards each half of the cell. In animals, the centrosome splits into two centrioles which move to the poles of the cell. The spindle seems to radiate from these two **centrioles**.

Metaphase

During this period, chromosomes become aligned at midpoint or **equator** between poles of the cell and are at their thickest and shortest structure. They are easily identified as two longitudinally double sister chromatids. In animals and plants, chromatids are connected (at their centromeres) to the spindle apparatus, which has formed between the two centrioles located at the poles of the cell. In many plants, the centrioles are absent. The spindle is still present, however, and the plant chromosomes are similarly attached to the spindle microtubular fibers.

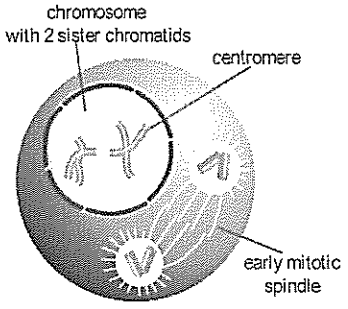


Figure 2: Prophase

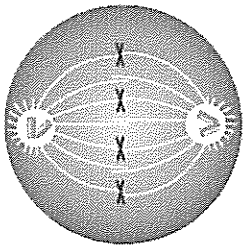


Figure 3: Metaphase

Background Information

Anaphase

In this short phase, sister chromatids begin to separate and migrate to the poles. Once the two chromatids separate, each is called a **chromosome**. For humans, with a diploid number of 46 chromosomes, there will be 46 chromosomes moving toward each pole. Onions have 16 **diploid** chromosomes and, therefore 16 chromosomes move to each pole. During anaphase there is a quantitative, equal segregation of the diploid number of chromosomes into two developing nuclei at the poles of the anaphase cell.

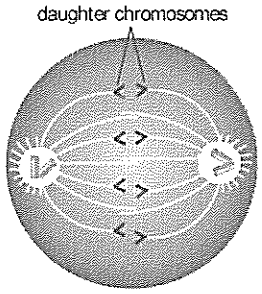


Figure 4: Anaphase

Telophase and Cytokinesis

The final mitotic phase of the cell cycle is recognized by the formation of two new nuclei encompassing the daughter chromosome at the cell poles. The mitotic apparatus disappears and chromosomes begin to lengthen as they unwind. **Cytokinesis**, formation of a new cell membrane, occurs midway between the daughter nuclei. In animals, there is the formation of the indented **cleavage furrow**. In plants, such as the onion root tip cells, this is seen as the formation of a cell plate, dividing the original cell into two (presumably equivalent) daughter cells. Cells now enter the G1, stage of interphase in the cell cycle and the process begins anew.

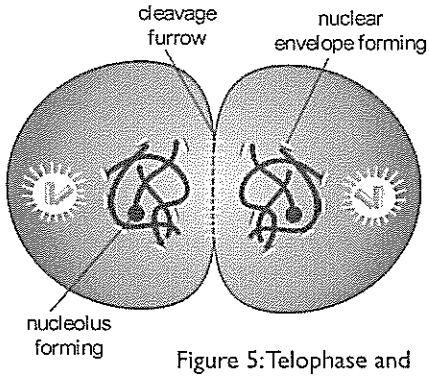


Figure 5:Telophase and Cytokinesis

Meiosis

Meiosis is a specialized type of cell division sharing many features with mitosis. The main difference is that meiosis involves two successive nuclear divisions that produces four **haploid** cells. Each gamete, or sex cell, contains half the number of chromosomes. In humans, each gamete contains 23 chromosomes. Fertilization of an egg by a sperm, each containing 23 chromosomes, restores the diploid number of 46 chromosomes. Meiosis consists of two rounds of cell division, **Meiosis I** and **Meiosis II**, each with its own prophase, metaphase, anaphase and telophase.

In animals, the gametes, sperm and egg of animals are generally formed directly from diploid tissue rather than from a haploid gametophyte generation in plants such as corn. In animals, the egg and sperm join to form the diploid zygote which develops into a mature adult. In plants, one of the male gametes from the pollen (formed in the stamens) unites with the female gamete in the pistil to form the fertilized diploid zygote. The other male gamete combines with the diploid endosperm nucleus to form a triploid endosperm tissue. Both are in the corn seed.

Background Information

MEIOTIC DIVISION I

Prophase I

The chromosomes begin to shorten and thicken. In some plants, they appear to aggregate together on one side of the nucleus. In animals, they may appear to orient with one end nearest the nuclear membrane adjacent to the centriole. **The first major difference between mitosis and meiosis is that homologous pairs of chromosomes come together or synapse.** A **tetrad** consisting of four chromatids is the result. This complex allows for "crossing over" to occur between the homologous pairs of chromosomes. The point of crossing over appears as an X shaped structure, called the **chiasma**, (**chiasmata**, plural). During the formation of the chiasmata, there is a crossing over, or genetic exchange, between homologous chromosomes. There is an enzyme catalyzed breakage and repair of the synapsed chromosomes. Crossing over is very important because it leads to an increase in genetic randomness and species/genetic diversity. The last step is the ending of chiasmata formation, disappearance of the nucleolus and nuclear membrane, and formation of the mitotic spindle.

Metaphase I

The synapsed homologous pairs of chromosomes arrive at the midpoint, or **equator**, between poles. The synapsed pairs orient such that one member of each pair faces the opposite pole of the cell, with the 23 pairs of chromosomes arranged entirely in random fashion. There is no tendency for one member of the pair to face one of the poles. This random assortment also contributes heavily to genetic diversity within a species.

Anaphase I

The pairs of homologous chromosomes, each longitudinally double (tetrads), begin to separate and migrate to the cell poles. As contrasted to mitosis, entire chromosomes, versus the sister chromatids, move to each pole. **This is the second major difference between mitosis and meiosis.** Each pole randomly receives either the maternal or paternal chromosome of each homologous pair. Therefore, there is an exact halving of the diploid chromosome number during Anaphase I stage of meiosis.

Telophase I

The chromosomes arrive at the poles of the cell at the beginning of this phase. The nuclear membrane forms and the nucleolus begins to reorganize. Cytokines is, physical cell division, occurs during this phase, although not in all animal or plant species. In corn, there is a physical separation during this stage. In the plant Trillium, Telophase I appears to be skipped entirely. Interphase II (Interkinesis). How much time spent in this phase depends on the type of organism, the formation of new nuclear envelopes, and the amount of chromosomal uncoiling. A third major difference between mitosis and meiosis is that DNA replication does not occur during interkinesis.

Background Information

MEIOTIC DIVISION II

In order to reduce the amount of DNA to half, a second meiotic division is necessary to separate the chromatids of the chromosomes in the two daughter cells formed in Meiosis #1.

Prophase I

This phase resembles mitotic prophase except the chromosomes do not dramatically shorten. The nucleolus, the site of active rRNA synthesis, disappears. The nuclear membrane also disappears and the mitotic apparatus, the spindle, begins to organize within the cell.

Metaphase II

The monoploid number of chromosomes organizes at the midpoint (equator) between the poles. Each chromosome is composed of two sister chromatids.

Anaphase II

The sister chromatids begin to separate and migrate to the poles as in mitosis. This stage ends when they are at the poles. Each chromatid has its own separate centromere region now, and it is called a chromosome.

Telophase II

The chromosomes begin to lengthen, the nucleus reforms, and the nucleolus reorganizes. Cytokinesis occurs and the final result of meiosis is four cells each containing the haploid chromosome number of chromosomes.

Meiosis, therefore, is a process that produces gamete diversity – independent assortment. This independent assortment provides chromosomes without crossover (Figure 6), and chromosomes with crossover (Figure 7). Chromosomal crossover (or crossing over) is the exchange of genetic material between homologous chromosomes that results in recombinant chromosomes. It is one of the final phases of genetic recombination, which occurs during prophase I of meiosis.

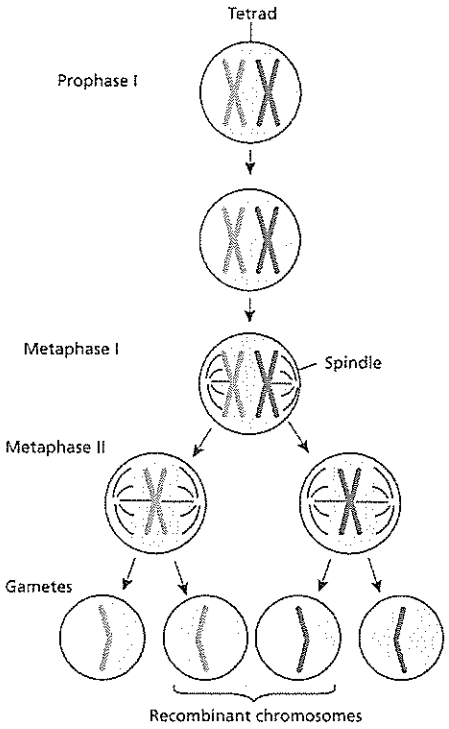
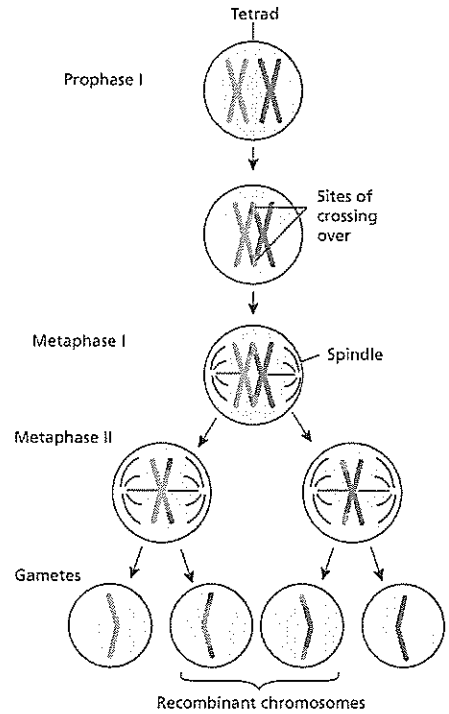


Figure 6 (above): Meiosis without crossing over
Figure 7 (below): Meiosis with crossing over



Background Information

CELL CYCLE AND CHECKPOINTS

Cell division is tightly controlled by complexes made of several specific proteins which contain enzymes called cyclin-dependent kinases (CDKs). CDKs turn on or off the various processes that take place in cell division. Cyclins are another family of proteins, with which CDK partners. For example, CDK is activated when it is bound to cyclin,interacting with various other proteins, and allow the cell to proceed from G2 into mitosis.

Cyclins and CDKs do not allow the cell to progress through its cycle automatically. There are three checkpoints a cell must pass through during its cycle: the G1 checkpoint, G2 checkpoint, and the M-spindle checkpoint (Figure 8). Cell cycle checkpoints are regulatory pathways that control the order and timing of cell cycle transitions and ensure that critical events such as DNA replication and chromosome segregation are completed with high fidelity. In addition, checkpoints respond to damage by arresting the cell cycle to provide time for repair and by inducing transcription of genes that facilitate repair. Checkpoint loss results in genomic instability and has been implicated in the evolution of normal cells into cancer cells

G1 (restriction) checkpoint is where the decision is made whether the cell will be divided, delayed division, or enter the resting stage. At **G2 checkpoint**, the success of DNA replication from the S phase is checked. If this checkpoint is passed, the cell initiates the many molecular processes that signal the beginning of mitosis. M checkpoint assures that the mitotic spindles or microtubules are properly attached to the kinetochores. If the spindles are not anchored properly, the cell does not continue on through mitosis. Mutations in cell cycle genes that interfere with proper cell cycle control are found very often in cancer cells.

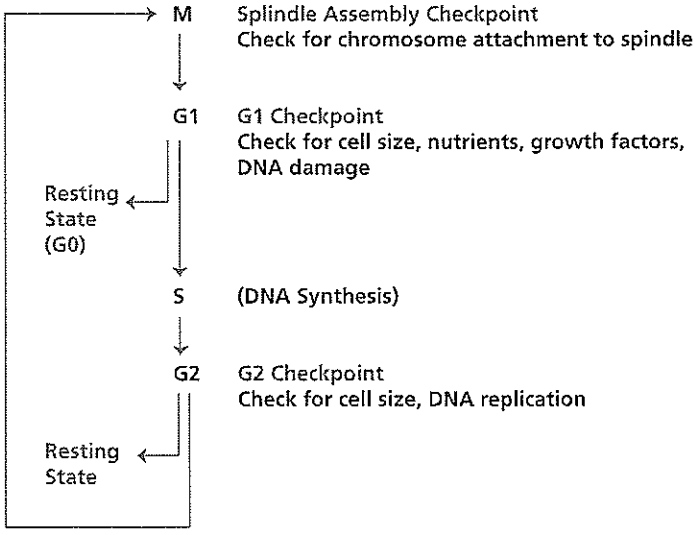


Figure 8: Cell Cycle with Checkpoints

Background Information

Human chromosomal disorders occur as a result of loss of control during cell cycle. Non-disjunction occurs when either homologues fail to separate during Anaphase I of meiosis, or sister chromatids fail to separate during anaphase II. The result is that one gamete has 2 copies of one chromosome and the other has no copy of that chromosome. (The other chromosomes are distributed normally.)

If either of these gametes unites with another during fertilization, the result is aneuploidy (abnormal chromosome number)

- A trisomic cell has one extra chromosome (2n +1) = example: trisomy 21. (Polyploidy refers to the condition of having three homologous chromosomes rather then two)
- A monosomic cell has one missing chromosome (2n - 1) = usually lethal except for one known in humans: Turner's syndrome (monosomy XO).

Patau syndrome is a chromosomal abnormality in which an individual has an additional chromosome 13 due to a nondisjunction of chromosome during meiosis. Down syndrome is a result of an extra copy of chromosome 21. People with Down syndrome are 47, 21+. Down syndrome affects 1:700 children and alters the child's phenotype either moderately or severely. Edward's syndrome is a genetic disorder in which a person has a third copy of material from chromosome 18, instead of the usual two copies.

MEIOSIS AND CROSSING OVER IN SORDARIA FIMICOLA

Sordaria fimicola (*S. fimicola*) offers great advantages for genetic studies as they have a short 7-12 day life-cycle, and are easily grown in culture. The most common form of *S. fimicola* is a dark brown while certain mutants are grey or tan. *S. fimicola* produces black perithecia containing asci. Each ascus contains eight ascospores a linear arrangement.

In the last investigation, students will discover how *S. fimicola* can give us information about crossing over during meiosis. If no crossing over, there is a 4:4 pattern - 4 black spores, and 4 tan spores all lined up. If crossing over does occur there is a 2:2:2:2 pattern visible, or a 2:4:2 pattern. Parental type asci have four black and four tan spores in a row (4:4 pattern), while recombinant asci will not have this pattern (Figure 9).

The frequency of crossing over appears to be governed largely by the distance between genes, or in this case, between the gene for spore coat color and the centromere. The probability of a crossover occurring between two particular genes on the same chromosome (linked genes) increases as the distance between those genes becomes larger. The frequency of crossover, therefore, appears to be directly proportional to the distance between genes.

A map unit is an arbitrary unit of measure used to describe relative distances between linked genes. The number of map units between two genes or between a gene and the centromere is equal to the percentage of recombinants.

Background Information

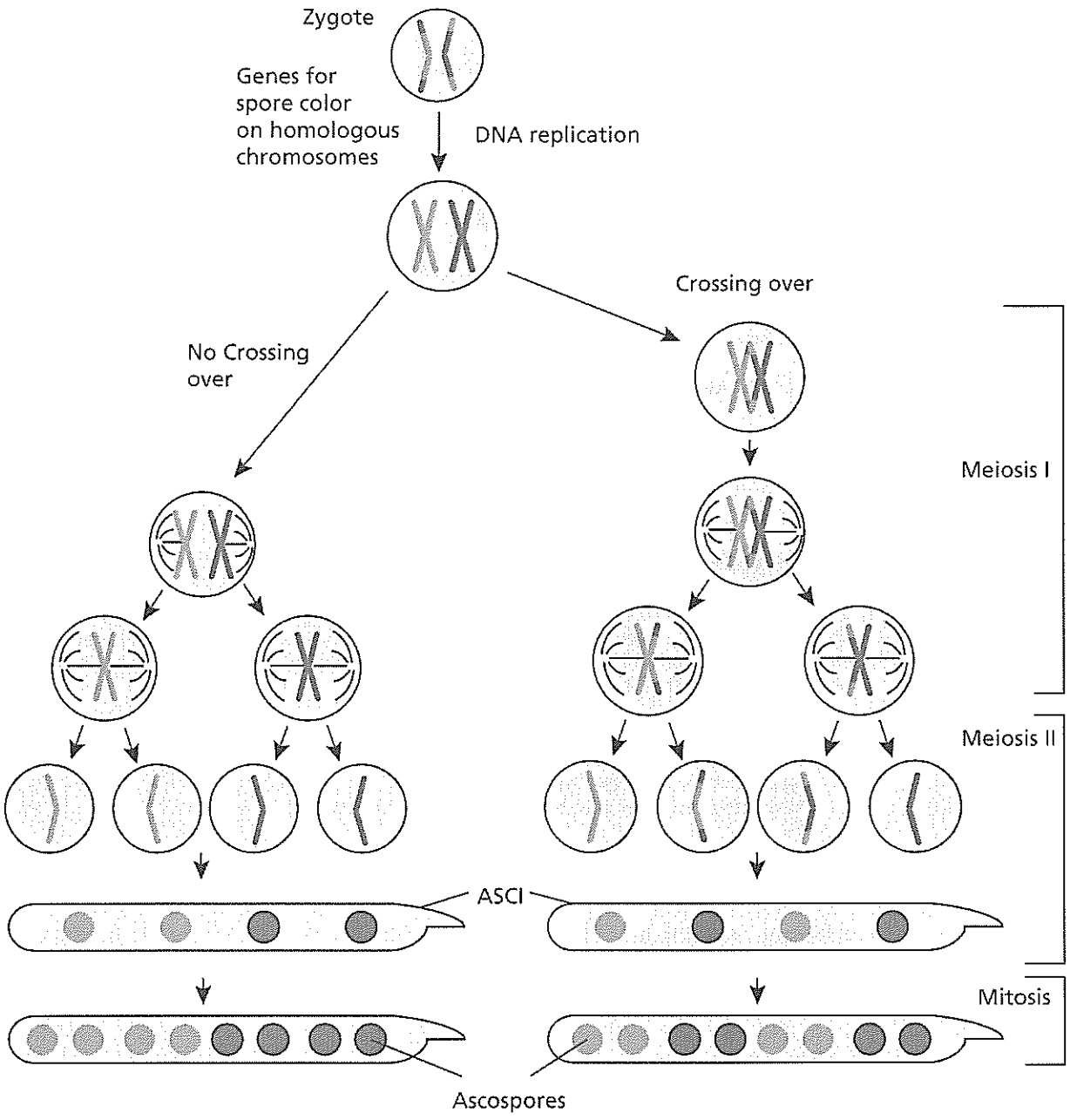


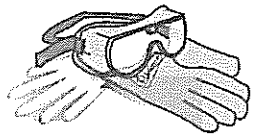
Figure 9: Meiosis and Crossing Over in Sordaria

Experiment Overview and General Instructions

EXPERIMENT OBJECTIVE:

The objective of this experiment is for students to identify and differentiate various stages in mitosis and meiosis. Onion root tips are stained to identify the various stages and duration of mitosis. Students will also have an opportunity to analyze mechanism involved with loss of cell cycle control in cancer. Meiosis and Crossing Over in *Sordaria* is also examined in this experiment.

LABORATORY SAFETY GUIDELINES



1. Wear gloves and goggles while working in the laboratory.
2. Exercise caution when working in the laboratory – you will be using equipment that can be dangerous if used incorrectly.
3. DO NOT MOUTH PIPET REAGENTS - USE PIPET PUMPS.
4. Always wash hands thoroughly with soap and water after working in the laboratory.
5. If you are unsure of something, ASK YOUR INSTRUCTOR!

LABORATORY NOTEBOOKS:

Scientists document everything that happens during an experiment, including experimental conditions, thoughts and observations while conducting the experiment, and, of course, any data collected. Today, you'll be documenting your experiment in a laboratory notebook or on a separate worksheet.

Before starting the Experiment:
- Carefully read the introduction and the protocol. Use this information to form a hypothesis for this experiment.
- Predict the results of your experiment.

During the Experiment:
- Record your observations.

After the Experiment:
- Interpret the results – does your data support or contradict your hypothesis?
- If you repeated this experiment, what would you change? Revise your hypothesis to reflect this change.

Investigation I & IV: Modeling Mitosis and Meiosis

Notes:

- This investigation is best done after students have at least read about mitosis and meiosis, and/or seen a video, or online animation showing these two processes. The activity is designed to help students learn the critical distinctions between what happens to chromosomes during mitosis vs meiosis.
- Teachers will provide guidance as students use pipe cleaners to review chromosome duplication and movement during mitosis and meiosis.

MITOSIS:

Students will work in groups to manipulate pipe-cleaner chromosomes on a template showing stages of mitosis with one pair of chromosomes. Place your model chromosome in each cell circle and manipulate the model chromosomes during the different stages of mitosis until approved by the teacher.

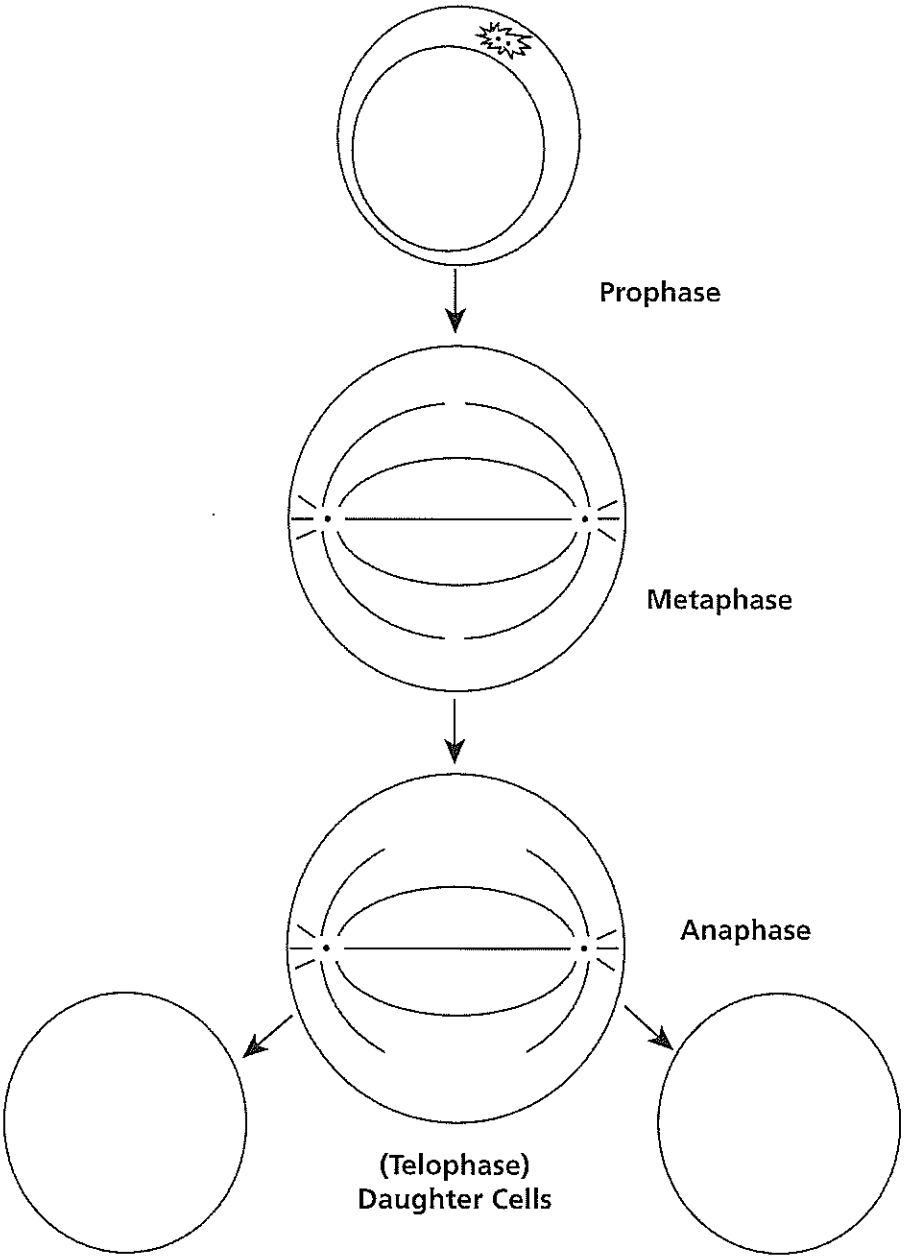
1. Each single fuzzy piece (pipe cleaner) equals one chromosome
   a. A piece of COLOR #1 pipe cleaner represents one chromosome inherited from the mother;
   b. A piece of COLOR #2 pipe cleaner represents one chromosome inherited from the father.
2. Two fuzzy pieces, held together by a bead—the centromere—represents one chromosome duplicated into two new strands (chromatids), each of which becomes a duplicate chromosome when the centromere splits at the beginning of anaphase.
3. Check your chromosome. Before doing this lab, AND when finished, count all pieces in the plastic bag. Notify your teacher if there are any extras or shortages. DO NOT REMOVE BEADS FROM DOUBLE FUZZY PIECES.
4. Arrange the pieces on Student Worksheet 1 - Mitosis, showing the essential chromosome arrangements during mitosis. You won't need all the pieces for this part. When done, raise your hand to be checked.
5. When your MITOSIS layout is approved, copy those arrangements onto Student Worksheet 1 – Mitosis, using two different color pencils.
6. Remove all pieces and proceed to arrange them on the two Student Worksheet 2 - Meiosis.

Remember:

- Prophase: The array of chromosomes in Prophase and the daughter cells is random, with the same two chromosomes in each cell.
- Metaphase: duplicated chromosomes (into chromatids, or "chromosome kids") are NOT paired off, but ARE lined up down the middle, in no particular sequence, top to bottom.
- Anaphase: The vertical sequence shown in metaphase should be followed with same sequence in Anaphase.

Investigation I & IV: Modeling Mitosis and Meiosis

STUDENT WORKSHEET 1: MITOSIS TEMPLATE



Investigation I & IV: Modeling Mitosis and Meiosis

MEIOSIS

Students will work in groups to manipulate pipe-cleaner chromosomes on a template showing stages of meiosis with one pair of chromosomes. Place your model chromosome in each cell circle and manipulate the model chromosomes during the different stages of meiosis until approved by the teacher.

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2. Two fuzzy pieces, held together by a bead—the centromere—equals one chromosome duplicated into two new strands (chromatids), each of which becomes a duplicate chromosome when the centromere splits at the beginning of anaphase.

3. Check your chromosome. Before doing this lab, AND when finished, count all pieces in the plastic bag. Notify your teacher if there are any extras or shortages. DO NOT REMOVE BEADS FROM DOUBLE FUZZY PIECES.

4. Arrange the pieces on Student Worksheet 2 – Meiosis, showing the essential chromosome arrangements during meiosis. You will need all the pieces for this part. When done, raise your hand to be checked.

5. When your Meiosis layout is approved, copy those arrangements onto Student Worksheet 2 – Meiosis, using two different color pencils.

6. As an extension, ask students to repeat the mitosis and meiosis process using two pairs of chromosomes. It is very important that students follow the vertical pathway consistently.

Remember:

1. Prophase: Array of chromosomes in Prophase is random.

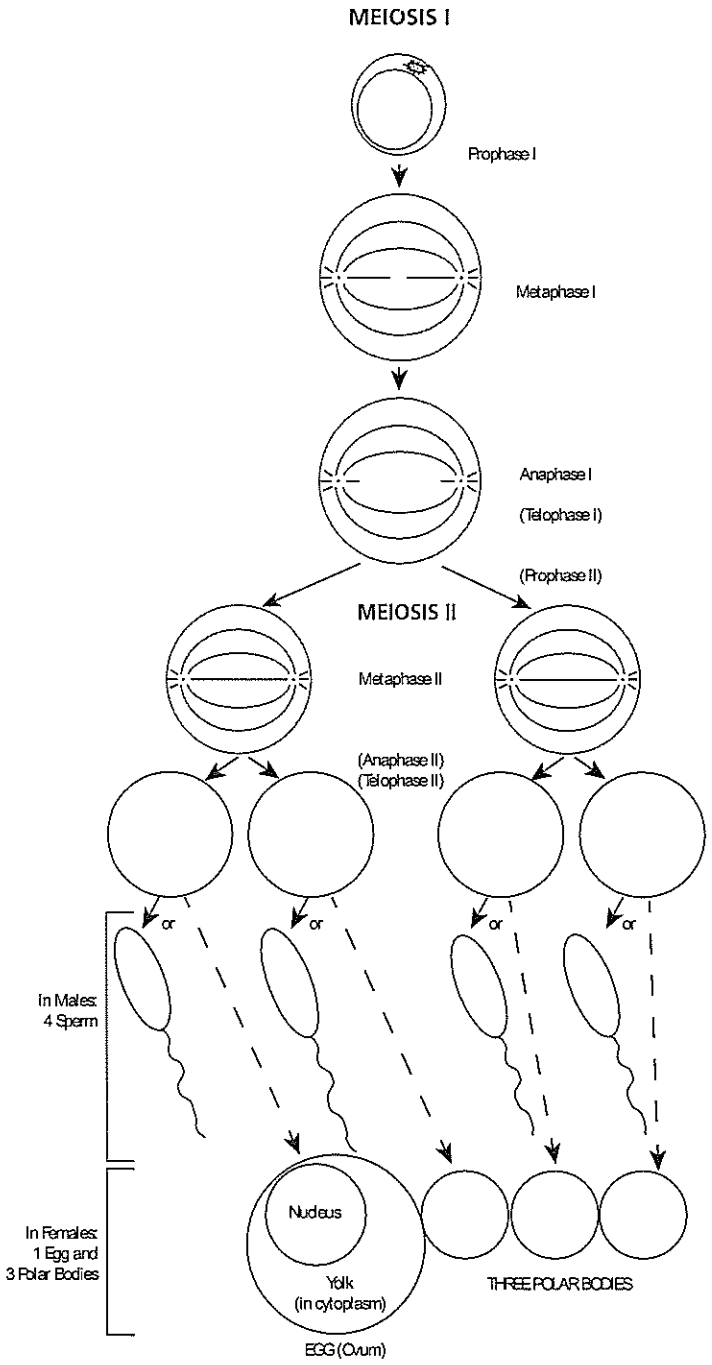
2. Metaphase I: duplicated chromosomes are paired (two longs are side by side, same for two shorts); vertical sequence can vary, likewise for which chromosome of each pair is on the left. For example, there can be a long green above a short red on the left, as shown, or a short red above a long red, etc. (Green and red are just two different colored pipe cleaners in our example.)

3. Anaphase I and Metaphase II: The vertical sequence shown in Anaphase I and Metaphase II should be followed with the same sequence in Metaphase I. This is important.

4. Males are to show chromosomes in sperm, females in eggs and polar bodies. Only one chromosome in each sex cell, with colors matching Metaphase II colors above.

Investigation I & IV: Modeling Mitosis and Meiosis

STUDENT WORKSHEET 2: MEIOSIS TEMPLATE



Experiment Procedure

Investigation II: Studying the Effects of Environment on Mitosis

You will prepare slides containing stained onion root tip squash sections, which will allow you to identify relevant stages of mitosis. Your instructor has prepared freshly rooted onions for use in this procedure. Remember it is the tips of newly emerging roots that contain the highest proportion of cells undergoing mitosis.

A. Preparation of the Chromosome Squashes:

1. Obtain the conical tube containing a newly rooted onion from your instructor.
2. Cut off approximately 1-2 mm of the root tip using a straight edged razor blade or a scalpel.
3. Add 5 ml of 12 M HCl to the conical tube. Transfer the onion root tip into the tube containing the HCl and incubate for 4 minutes.
4. Discard the HCl. Rinse the tube with distilled or deionized water.
5. Add 5 ml of Carnoy's solution to the tube. Transfer the tip to the Carnoy's fixative solution for 4 minutes.
6. Quickly rinse the slides with 70% Ethanol and dry it with a Kimwipe.
7. Place the onion tip on the slide, and cut off the distal 1-2 mm portion of the tips. Discard the remainder of the onion tip.
8. Add approximately 50 µl of Carbol Fuschin stain to cover the onion root tip. Leave the stain on the onion root tip for 2 minutes.
9. Blot off the excess stain with a Kimwipe.
10. Cover the tip with 1 – 2 drops of distilled or deionized water.
11. Place the cover slip over the tip.
12. Carefully and gently apply pressure on top of the cover slip. This will spread out the stained onion root tip for visualization.

Investigation II: Studying the Effects of Environment on Mitosis

B. Counting Cells and Analyzing Data:

1. As with all attempts to visualize material under the microscope you should begin at the lowest lens power that will visualize the objects of interest. Then shift to higher lens powers for further viewing.

2. Sketch the phases you observe. You should be able to identify all of the mitotic stages including: prophase, metaphase, anaphase, telophase and the nondividing stage, interphase. Record the number of cells in each stage.

3. Record the number of cells in each stage. Count at least three full fields of view. You should have counted over 200 cells.

4. Record your data in Table 1, below.

5. Calculate the percentage of cells in each phase and record in the table below. Estimate time spent in each stage by the following calculation:

   Number of cells in stage
   Total # cells in all stages

6. Calculate the amount of time spent in each phase of the cell cycle from the percentage of cells in that stage. On the average, it takes 1,440 minutes (24 hours) for onion root tip cells to complete the cell cycle.

   % of cells in stage X1440 minutes = _____ minutes of cell cycle spent in stage.

Table I	Number of Cells				Percent of Total Cells Counted	Time in Each Stage
	Field 1	Field 2	Field 3	Total		
Interphase						
Prophase						
Metaphase						
Anaphase						
Telophase						
	Total Cells Counted					

Experiment Procedure

Investigation III - Loss of Cell Cycle Control in Cancer

Figures 10 and 11 are pictures of 46 human chromosomes in a somatic cell, arrested in metaphase. Can you see that they are duplicated sister chromatids?

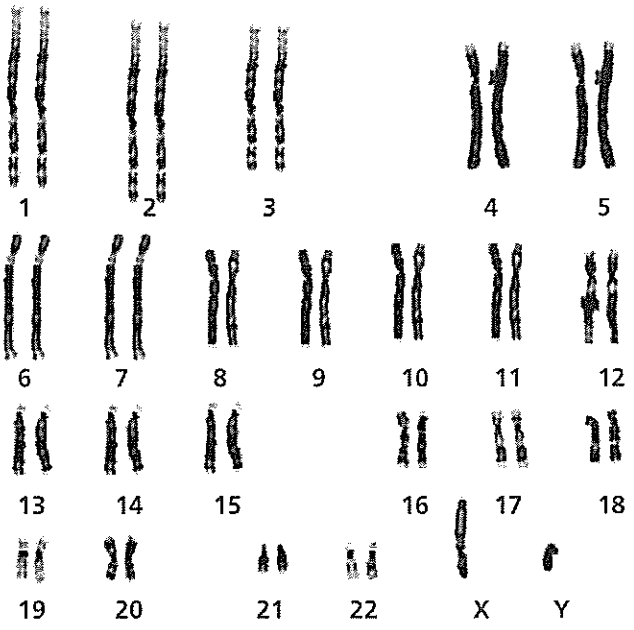


Figure 10: Normal Male Karyotype: 46, XY

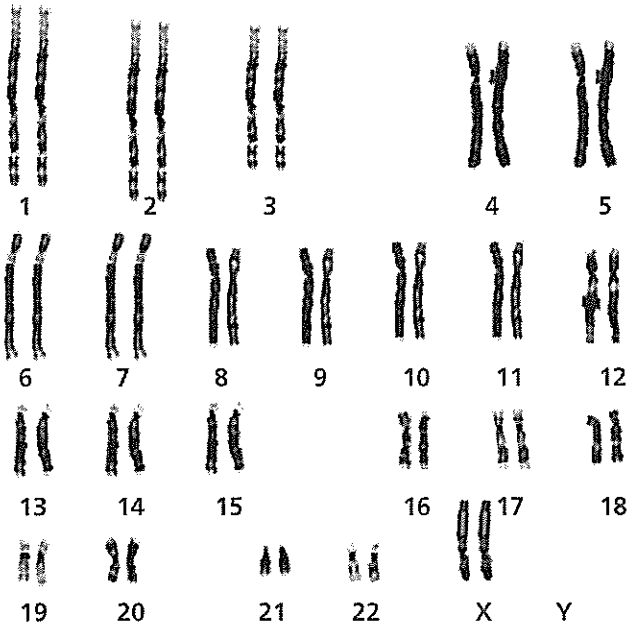
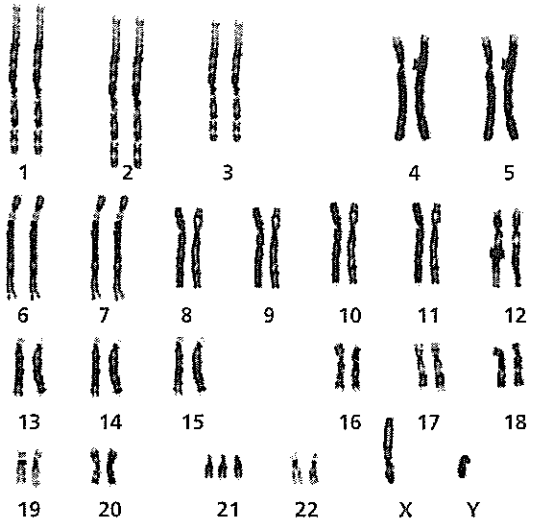


Figure 11: Normal Female Karyotype: 46, XX

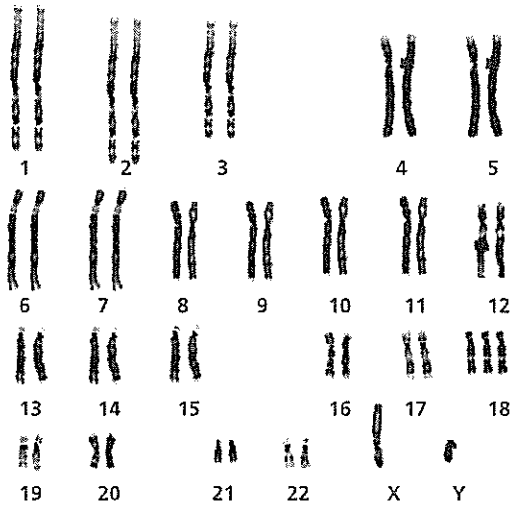
Investigation III - Loss of Cell Cycle Control in Cancer

STUDENT WORKSHEET 3: IDENTIFICATION OF KARYOTYPES FROM PATIENTS WITH CHROMOSOMAL DISORDERS

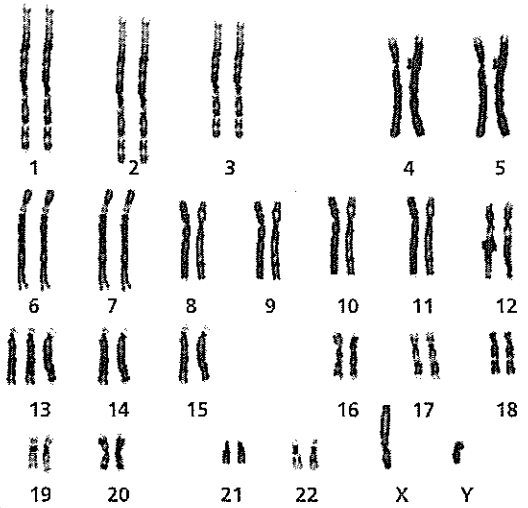
Based on your knowledge about human chromosomal disorders and nondisjunction due to loss of control during cell cycle, identify the name of the syndromes and karyotypes of the following patients.



Karyotype of Patient #1
Identify Syndrome:
Karyotype:



Karyotype of Patient #2
Identify Syndrome:
Karyotype:



Karyotype of Patient #3
Identify Syndrome:
Karyotype:

Investigation V: Meiosis and Crossing Over in Sordaria

In this investigation, students will measure crossover frequencies and genetic outcomes in a fungus. Your students will examine *Sordaria fimicola* asci produced by crossing wild type (black) with tan parents.

Each ascus contains eight spores. Parental type asci have four black and four tan spores in a row (4:4 pattern). Recombinant asci will not have this pattern.

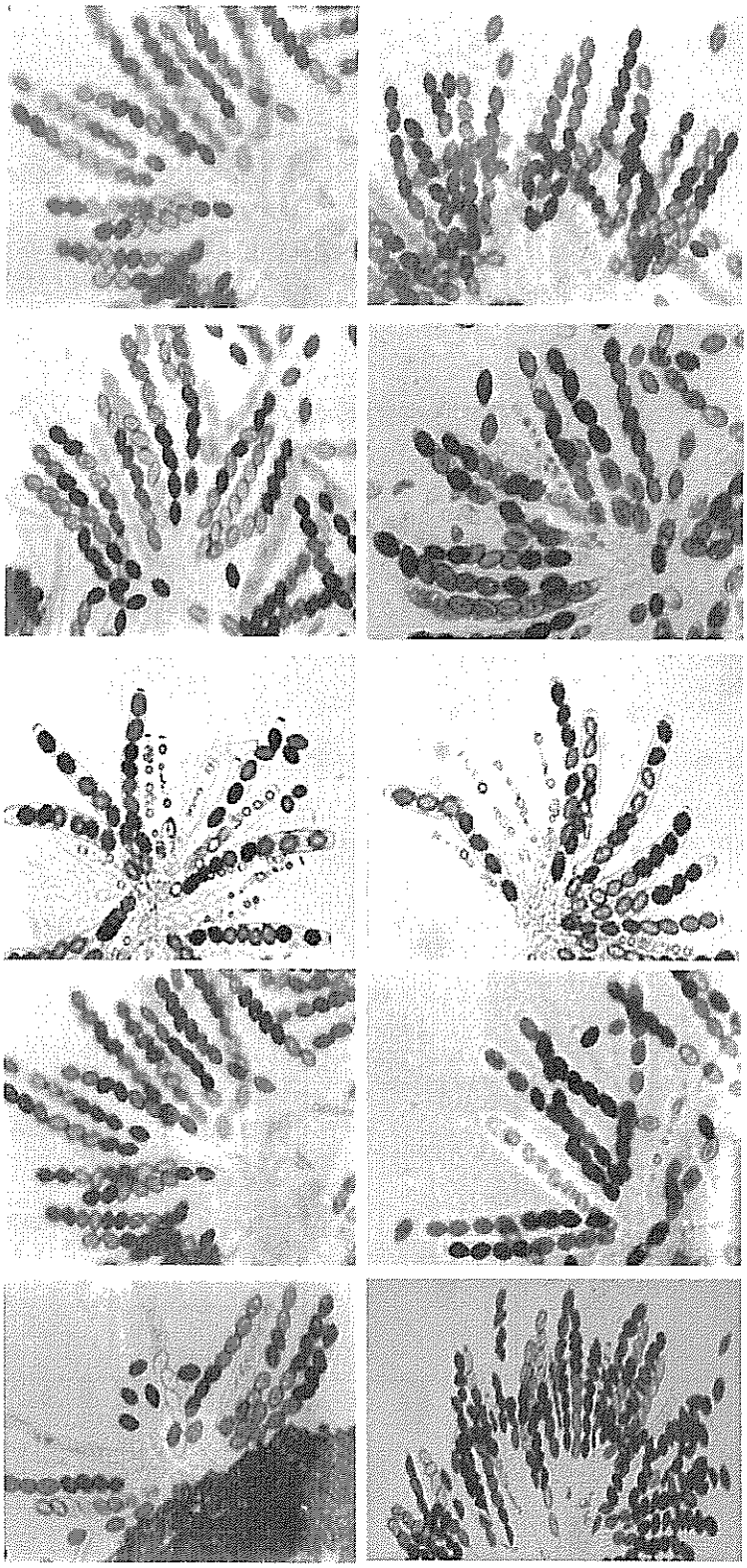
Study the pictures of Sordaria in Student Worksheet 4 provided by your lab instructor by counting at least 50 asci and scoring them as either parental or recombinant.

- If the ascospores are arranged 4 dark/4 light, count the ascus as "No crossing over."
- If the arrangement of ascospores is in any other combination, count it as "Crossing over."
- Record your result in table on Student Worksheet 4.

STUDENT WORKSHEET 4: IDENTIFICATION RECOMGINANT ASCI AND PARENTAL TYPES

1. Once you have determined if crossing over has occurred in at least 50 hybrid asci, record your data in table below.
2. Based on your counts, determine the percentage of asci showing crossover. Record in table below.
3. Divide the percent showing crossover by 2. This is your gene to centromere distance. (The percentage of crossover asci is divided by 2 because only half of the spores in each ascus are the result of a crossover event.)

Number Asci not showing Crossover	Number of Asci showing Crossover	Total	% Asci Showing Crossover	Gene to Centromere Distance (map units)



Study Questions

Answer the following study questions in your laboratory notebook or on a separate worksheet.

1. What is the significance of the "S" phase of Interphase of cell division?
2. What are the specific differences between animal and plant karyokinesis and cytokinesis?
3. Based on the number of cells you found in each stage of mitosis in the onion root tip, what stage of mitosis is the longest?
4. List and explain the 3 major differences between mitosis and meiosis during Prophase I, Anaphase I and Interphase II.
5. Compare mitosis and meiosis by completing the following table:

	Mitosis	Meiosis
Parent Cell Chromosome Number		
Number of DNA Replications		
Number of Cell Divisions		
Number of Daughter Cells Produced		
Daughter Cell Chromosome Number		
Major Significance		

6. What is the major difference between Meiosis I and Meiosis II?
7. When does crossing over usually occur? Why is this significant?
8. How does the frequency of crossing over relate to the distance between genes?
9. Why is it important to know the percentage of recombinants in offspring?
10. What is a map unit?

Notes: