

LAB 9 – EUKARYOTIC CELL DIVISION: MITOSIS AND MEIOSIS

Objectives

1. Identify plant and animal cells in each stage of mitosis.
2. Model each stage of mitosis and meiosis.
3. Assess the generation of genetic diversity due to the independent assortment of chromosomes.

INTRODUCTION

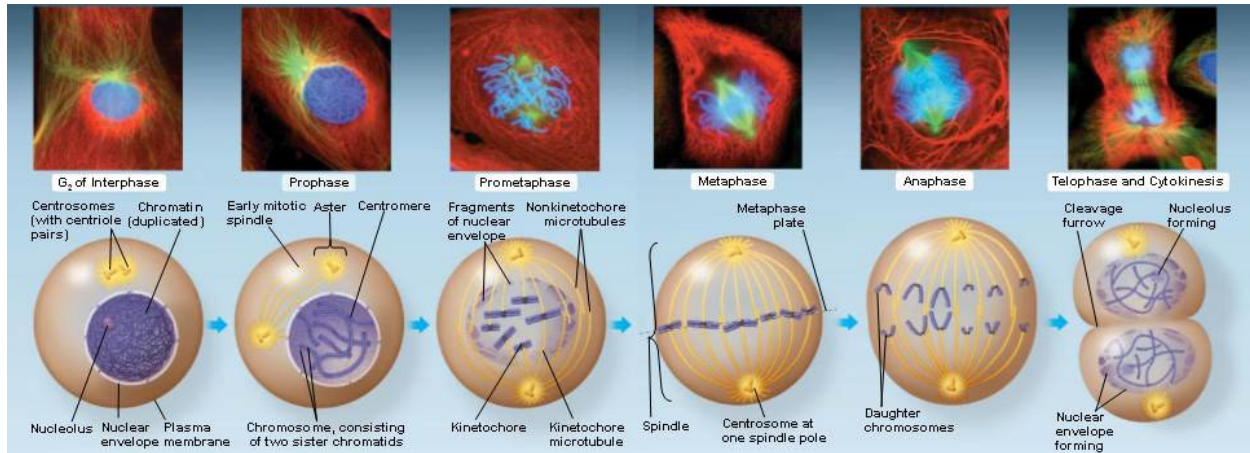
BINARY FISSION: Prokaryotic cells (bacteria) reproduce asexually by binary fission. Bacterial cells have a single circular chromosome, which is not enclosed by a nuclear envelope. During binary fission the bacterial chromosome is duplicated, the cell elongates, and the two chromosomes migrate to opposite ends of the cell. Each daughter cell receives one chromosome and is identical to the parent cell. Binary fission is a relatively fast and simple process.

MITOSIS: The increased complexity of eukaryotic cells causes several logistical problems during cell division. Eukaryotes are **diploid**, which means they have **two** sets of chromosomes; one set of chromosomes is inherited from each parent. Eukaryotic DNA is enclosed by a nuclear envelope. The proper sorting and distribution of multiple chromosomes during cell division is a complex process that requires the temporary dissolution of the nuclear envelope. Eukaryotic organisms carry out **mitosis** throughout their entire life to grow and to replace old or damaged cells. Some eukaryotic organisms use mitosis to reproduce asexually. The daughter cells produced by mitosis are **diploid** and genetically identical to each other and the parent cells that produced them.

CELL CYCLE: INTERPHASE & MITOSIS

Cells only spend a small part of their life dividing. The time between consecutive mitotic divisions is referred to as **interphase**. Eukaryotic cells spend most of their time in interphase. During interphase the cell's genetic material is in the form of chromatin (uncoiled DNA), nucleoli are present, and the nuclear envelope is clearly visible. Shortly before mitosis, the cell duplicates its DNA during the **S (synthesis) phase** of interphase.

Mitosis can be divided into distinct phases as shown below:



I. Prophase: Nuclear envelope and nucleoli disappear. Chromatin condenses into **chromosomes**, which are made up of two identical **sister chromatids** joined by a **centromere**. In animal cells, **centrioles** start migrating to opposite ends of the cell (centrioles are not present in plant cells). The mitotic spindle forms and begins to move chromosomes towards the center of the cell.

NOTE: Prometaphase is a transitional phase between prophase and metaphase.

II. Metaphase: Brief stage in which chromosomes line up in the equatorial plane of the cell. In animal cells, one pair of centrioles are visible at both ends of the cell. The mitotic spindle is fully formed.

III. Anaphase: Sister chromatids begin to separate, becoming individual chromosomes, which begin to migrate to opposite ends of the cell.

IV. Telophase: A full set of chromosomes reaches each pole of the cell. The mitotic spindle begins to disappear. The nucleus and nucleoli begin to reappear. Chromosomes begin to unravel into chromatin.

Cytokinesis or **cytoplasmic division** usually occurs at the end of telophase. In plant cells cytokinesis is accomplished by the formation of a **cell plate**. Animal cells separate by forming a **cleavage furrow**. In some cells (e.g., muscle cells and certain embryonic cells) cytokinesis does not occur or is delayed until multiple nuclear divisions have occurred. So although cytokinesis is associated with telophase of mitosis, it is separate from mitosis and may not occur at all. Mitosis is therefore the process of **nuclear division**, distinct from cytokinesis or cytoplasmic division.

EXERCISE 1 – Observing mitosis under the microscope

Examine prepared slides of both plant cells (onion/allium root tip) and animal cells (whitefish blastula) under the microscope at 400X. Even though the cells in these tissues are rapidly dividing, **most** of the cells you see will be in **interphase** (between cell divisions). Using your microscope, scan the slides to **find a cell in interphase and each one of the four stages of mitosis**. Draw a schematic representation of your observations for **both plant and animal cells** at each stage in the spaces provided on your worksheet and be sure to indicate and clearly label the important features or events of each stage.

MEIOSIS

During sexual reproduction in eukaryotes, a **haploid sperm** cell fuses with a **haploid egg** cell to produce a **diploid zygote** or fertilized egg. In most species, it is very important that the offspring produced by fertilization have the same number of chromosomes as the parents. Even a single extra or missing chromosome can be lethal or extremely deleterious to an individual (e.g., Down's syndrome in humans). Meiosis is a special type of cell division that produces haploid gametes (sperm cells or ova). Meiosis only occurs in an individual's gonads, during their reproductive years.

Meiosis involves two cell divisions and ultimately produces four haploid gametes. The haploid gametes produced by meiosis are different from each other as well as from the parent cells due to the **crossing over** of genetic material between homologous chromosomes and the **random distribution** of homologous chromosomes.

Meiosis is different in males and females:

Spermatogenesis

- In males four functional sperm cells are produced by meiosis.

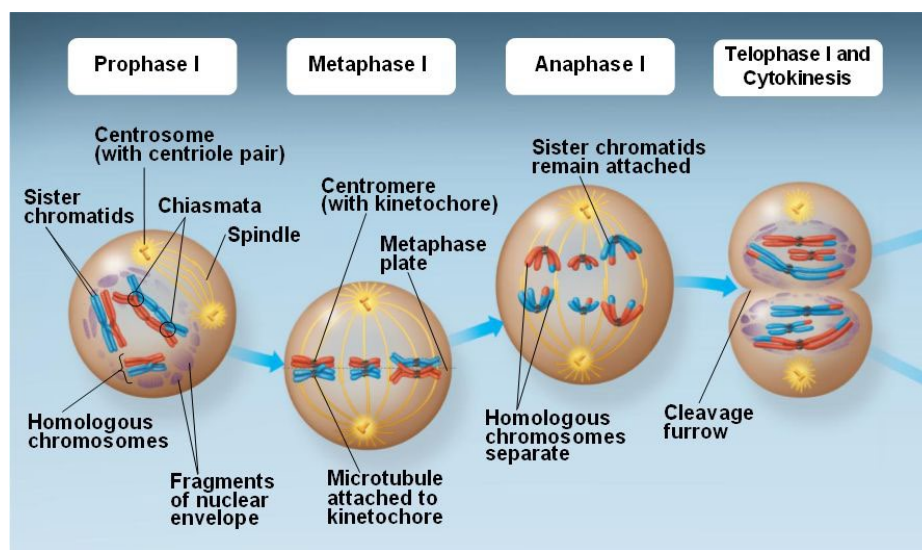
Oogenesis

- Due to unequal distribution of cytoplasm in during meiosis, one large functional egg (ovum) and three small **polar bodies** are produced.

STAGES OF MEIOSIS

MEIOSIS I

This is a reductive division in which one diploid (2N) cell produces two haploid (1N) cells.



Prophase I: Similar to prophase of mitosis with one important difference: **Crossing Over** in which pairs of **homologous** chromosomes **synapse** together to form **tetrads** and exchange genetic information (DNA). Crossing over creates new, recombinant chromosomes. Homologous chromosomes contain the same arrangement of genes and are of the same size. Although they are very similar (hence the term “homologous”), they differ slightly in DNA sequence since one comes from an individual’s mother, and the other comes from an individual’s father.

Metaphase I: Brief stage in which **tetrads** line up in the equatorial plane of the cell.

Anaphase I: Homologous chromosomes separate and migrate to opposite ends of cell. Sister chromatids DO NOT separate.

Telophase I/Cytokinesis: A full set of chromosomes reaches each pole of the cell. The cells produced contain half of the original number of chromosomes and are considered **haploid**. Each chromosome consists of two sister chromatids attached at the centromere.

Interphase may be very brief or absent between meiosis I and meiosis II.

MEIOSIS II

This division is very similar to mitosis – sister chromatids are distributed into different cells.

Prophase II: DNA condenses into chromosomes. No crossing over occurs.

Metaphase II: *Individual* duplicated chromosomes line up in the equatorial plane of the cell.

Anaphase II: Sister chromatids separate and begin to migrate to opposite poles of the cell.

Telophase II/Cytokinesis: A full set of chromosomes reaches each pole of the cell. Four genetically unique gametes are produced.

Exercise 2A – Modeling the stages of meiosis

1. Use the chromosome bead models to construct a single pair of **homologous chromosomes**, each with two sister chromatids. Use red for the maternal chromosome and yellow for paternal chromosome. Use the rubber tubing with magnets for centromeres and attach 10 beads to each end of the centromere.
2. Model each stage of meiosis with your single pair of homologous chromosomes, diagramming each stage on your worksheet using different colors to indicate maternal and paternal chromosomes. Also, be sure to include one crossover event during Prophase I.

INDEPENDENT ASSORTMENT OF CHROMOSOMES

For each homologous pair of chromosomes, maternal and paternal chromosomes are randomly distributed into daughter cells during meiosis I. The chromosomes of each homologous pair are distributed independently of the other homologous pairs, a phenomenon we refer to as **independent assortment**. Independent assortment can create a staggering number of possible chromosome combinations in gametes depending on the haploid chromosome number (n).

The number of possible gametes generated by independent assortment alone (i.e., without considering the effects of crossing over) is 2^n , where n is the haploid number of chromosomes in a cell. For example, in *Homo sapiens* (humans) $n = 23$ thus a human cell can produce over 8 million different gametes by independent assortment ($2^{23} = 8.4$ million). The effect of crossing over further increases genetic variation in gametes generated by an individual such that the number of possibilities is essentially infinite.

Exercise 2B – Modeling independent assortment

Use the chromosome bead models to model meiosis with one, two, or three pairs of homologous chromosomes. By doing this exercise you should be able to determine all the different combinations of chromosomes that are possible in gametes due to the independent assortment of homologous pairs. Diagram all possible arrangements in the circles provided using red for maternal chromosomes and yellow for paternal chromosomes.

NOTE: You will disregard crossing over between homologous chromosomes for this exercise.

Before you leave, please make sure your table is clean, organized, and contains all supplies listed below so that the next lab will be ready to begin. Thank you!

Lab Supply List

- Lens paper
- Chromosome bead kit

Also, please attend to the following before you leave:

- Return all **prepared** slides to the correct trays.
- Put the compound microscopes away as indicated below, and **make sure there is no slide on the stage:**
 - turn off power
 - secure the power cord to the back of the microscope
 - set to the lowest magnification (4X objective)
 - cover and place “arm out” in the cupboard at your table

LABORATORY 9 – WORKSHEET

Name _____

Section _____

Ex. 1 – Observing mitosis under the microscope

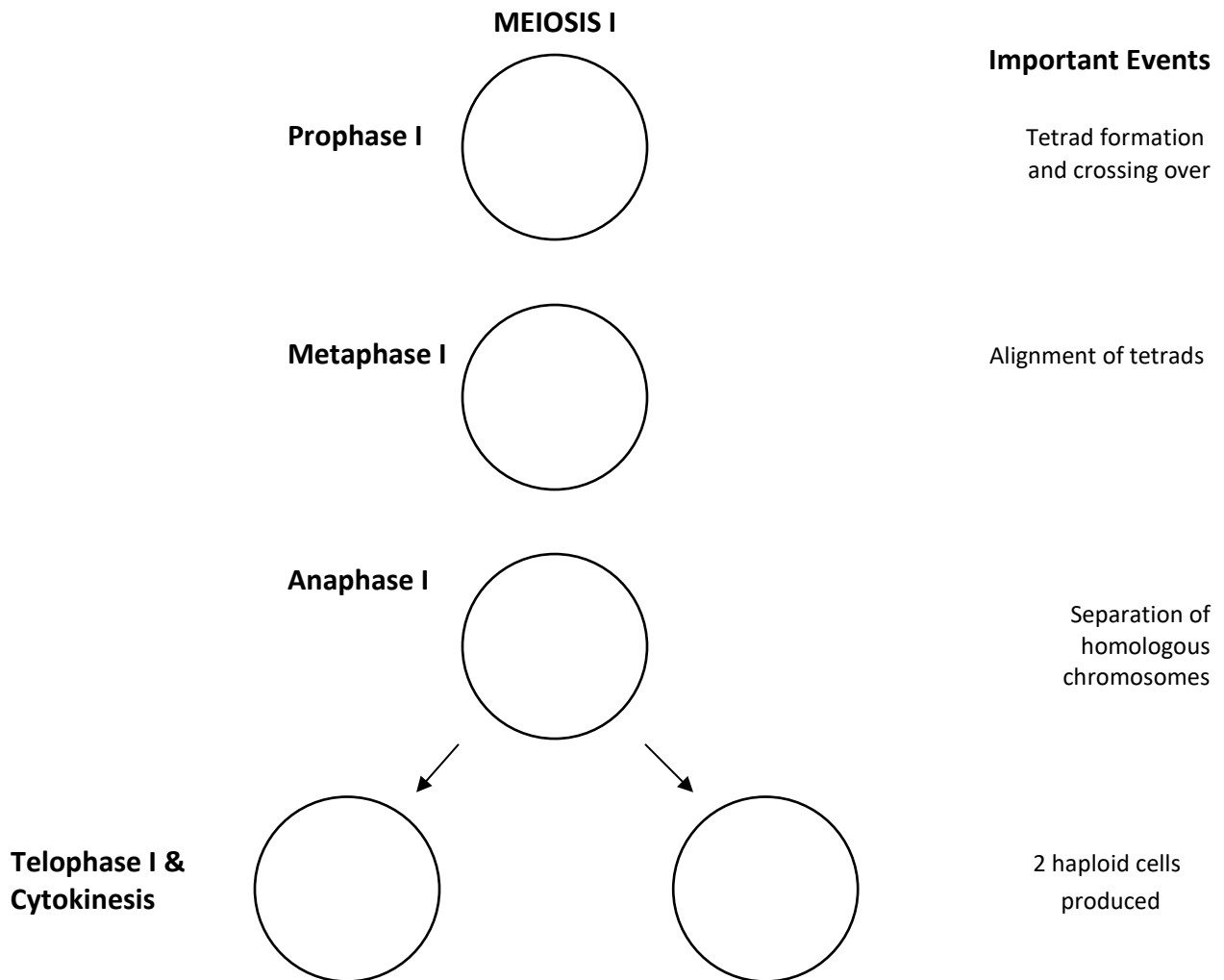
Diagram a single cell in each of the indicated stages of mitosis both *Onion Root Tip (plant)* and *Whitefish Blastula (animal)*:

ONION ROOT TIP (<i>Allium</i>)	WHITEFISH BLASTULA
Interphase	Interphase
Prophase	Prophase
Metaphase	Metaphase
Anaphase	Anaphase

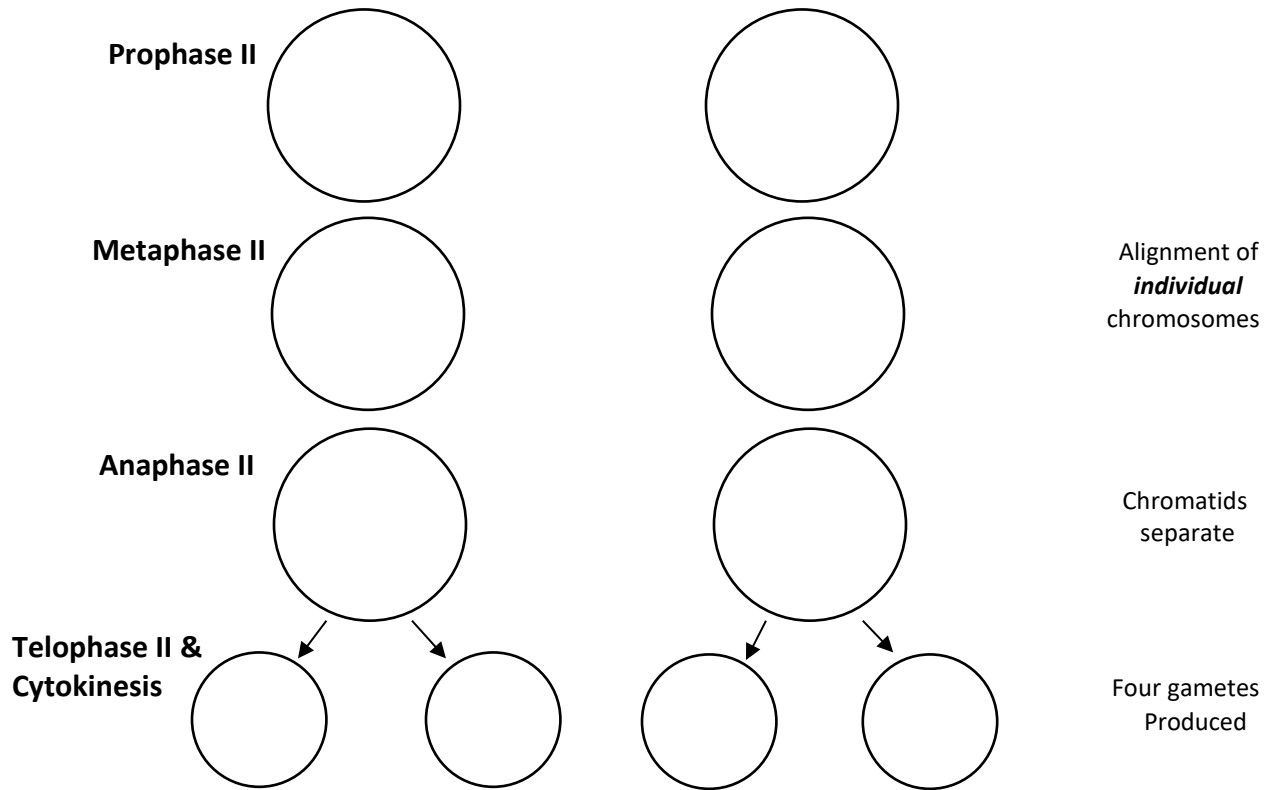
ONION ROOT TIP (<i>Allium</i>)	WHITEFISH BLASTULA
Telophase	Telophase

Ex. 2A – Modeling the stages of meiosis

Diagram each stage of meiosis as it unfolds for a cell with 1 pair of homologous chromosomes. Be sure to use different colors for maternal and paternal chromosomes and to have one crossover event.



MEIOSIS II

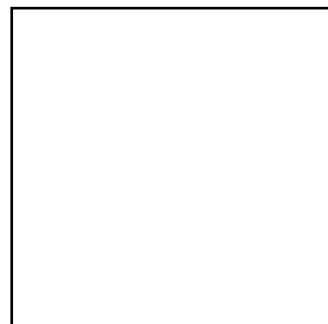
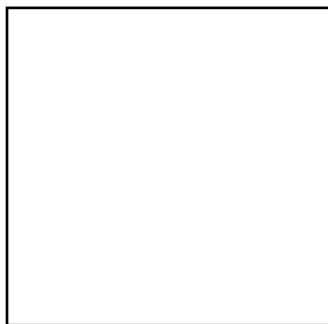


Ex. 2B – Modeling independent assortment

1. A cell with 1 pair of homologous chromosomes ($n = 1$)

Assemble two duplicated chromosomes, one red and one yellow, with 6 beads on each end of the centromere. By modeling meiosis, determine all possible combinations of chromosomes in gametes when $n = 1$, **assuming there is NO crossing over**, and diagram each **unique** combination in the squares below using different colors for maternal and paternal chromosomes.

$2^1 = \underline{\hspace{2cm}}$ (possible chromosome combinations when $n = 1$)



2. A cell with 2 pairs of homologous chromosomes ($n = 2$)

Assemble another homologous pair of duplicated chromosomes with 4 beads on each side of the centromere. Use this homologous pair with the longer homologous pair from the previous exercise to model meiosis when $n = 2$ and determine all possible combinations of chromosomes in gametes, **assuming there is no crossing over**. Diagram each **unique** combination in the squares below using different colors for maternal and paternal chromosomes.

$2^2 =$ _____ (possible chromosome combinations when $n = 2$)

--	--	--	--

3. A cell with 3 pairs of homologous chromosomes ($n = 3$)

Assemble another homologous pair of duplicated chromosomes with 2 beads on each side of the centromere. Use this homologous pair along with the homologous pairs from the previous exercise to model meiosis when $n = 3$ and determine all possible combinations of chromosomes in gametes, **assuming there is no crossing over**. Diagram each unique combination in the squares below using different colors for maternal and paternal chromosomes.

$2^3 =$ _____ (possible chromosome combinations when $n = 3$)

Question: How many gametes would be produced by independent assortment alone, in a cell with 8 pairs of homologous chromosomes? _____