LAB 10 Meiosis and Development (May 2014)

Section 1 – Mitosis and Meiosis Compared

[2] Yes, we've reached that point in the Biology 3 course to discuss the birds and the bees. Personally, I've never really understood that euphemism – humans don't have sex like birds or bees, let me assure you! But one characteristic that all the sexually reproducing organisms do share is the need to produce some special cells when it comes time to reproduce.

[3] Remember last week when we discussed mitosis – the reproduction of body, or somatic cells? The cells started off as diploid and produced two identical diploid cells. Almost every cell in your body possesses the same complement of chromosomes – 22 autosomes and a pair of sex chromosomes. It is critical that your somatic cells have a full complement of two sets of chromosomes to function normally. Answer the review questions regarding mitosis before we go on.

[4] Today however, we'll look at the process of producing some rare cells in your body – the sex cells or gametes. This process is called meiosis and although we will be using all the same cellular structures as we saw in mitosis, the resulting cells will be very different indeed. Record the definition of meiosis, then answer the next few questions in the lab as we go along.

[5] The cells produced by meiosis are produced only in the gonads – either the ovaries or testes. We'll look at these specialized sex organs later in the lab, because it turns out that the production of eggs and sperm are quite different given their very different roles in reproduction.

[6] As you know, the sperm and egg will eventually come together, so this involves making sure that the number of chromosomes is maintained for each species. How is this number maintained? All we have to do is make sure that the gametes have just **one** set of chromosomes, so that when fertilization happens, the resulting zygote gets back to the full diploid number. To abbreviate the chromosome numbers, we'll use the symbols shown here -1N for haploid, 2N for diploid.

[7] Think for a moment what would happen if organisms didn't produce haploid gametes: every generation would be doubling the number of chromosomes in their cells. For humans, if we started with 46 chromosomes, the kids would get 92, the grandkids get 184 and so on. Cellular reactions would get very confused very quickly.

[8] Take a look at these two cellular reproduction patterns side by side. As we saw last week, mitosis started with a diploid cell and ended with two genetically identical diploid cells. Meiosis will start with the same diploid cell, but through some fancy chromosome moves, end up with just half the number of chromosomes as the original cell. This is accomplished with two successive divisions – a very different process from mitosis.

[9] What about organisms that reproduce asexually? Organisms such as this *Paramecium* can use mitosis to produce a new individual, but the new organism would be genetically identical to the original with no chance of variation between individuals, and honestly, where's the fun in that?

[10] Sexual reproduction, however insures that the offspring are different than either of their parents. This is because of those haploid gametes produced in the gonads – only half the genes are being passed on from either parent, and those genes are combined with another individual's genes through fertilization to produce that new unique individual. Having two parents means new combinations in every generation and this is great benefit to species in the long term.

[11] There is a progression of these two types of cell reproduction. Meiosis produces the haploid gametes, the gametes join to form a diploid zygote, then the zygote grows into an adult organism, which then starts producing the haploid gametes with meiosis once again. Think of the proverbial chicken and egg question – as long as we think of it being a cycle, we won't lose too much sleep over the answer.

[12] Study the differences in the summary diagram in your book and answer the questions below the diagrams before you continue.

[13] Before we jump into the process of meiosis, there are certain chromosomes that we'll be on the lookout for, because they will interact in a way we did not see during mitosis. These chromosomes are called homologous chromosomes. Please draw these chromosomes and record the definition before we go on.

[14] Remember the pairings of chromosomes you saw last week in the karyotypes? Diploid organisms such as humans have two sets of chromosomes, one set from each parent, and we can pair them up because of their similar size and shape. These chromosomes carry the same *kind* of genetic information, but by no means *identical* information.

[15] Here is one of the pairs, chromosome pair number eight, pulled out from a human karyotype. We can't actually tell by looking at the chromosomes which came from the father and which came from the mother, but we can tell from the banding pattern that they are similar.

[16] Although the chromosomes of a homologous pair look alike, they wouldn't be alike in terms of specific genetic information because they reflect the information contributed by your mother **and** your father. Both your parents gave you information for hair color, eye color, blood type, and every other gene you inherited, but not necessarily the exact same genes. We'll assign the labels of maternal and paternal to keep them straight during today's lab. Record the definitions before you continue.

[17] Now you're probably one step ahead of me and already asking about those sex chromosomes... In males the two sex chromosomes, the X and the Y, are very different in size and shape. We still consider them a homologous pair because they carry the same type of information – the information for the development of sex characteristics. Complete the questions at the end of Section 1.

Section 2 – Stages of Meiosis

[18] Let's take a look at the process of meiosis all the way through. As we go, you will be coloring in the chromosomes as they appear on the program with the red and yellow pencils provided in your booth, then you'll have a nice visual summary when it comes time to try the chromosome model for

yourself. In a later part of the lab, after you've physically moved the chromosome models around, you'll be demonstrating the whole process to the lab instructor. This may sound scary, but we will practice several times, so don't panic. Yet.

[19] Okay, take a look at the summary in Section 2 of your lab book. Remember throughout this process that our goal is to get haploid gametes that will have a single set of chromosomes. If we wanted to look at human meiosis, we would have to draw 46 chromosomes to start, so let's not do that and just summarize the process in a hypothetical organism with just four chromosomes as its diploid number.

[20] This first drawing represents a diploid cell in a reproductive organ. Drag the labels to the proper part of the cell, then record these labels on the drawing at the top of Section 2 in your book when you get them all correct. Now the fun part - color in the cell to reflect the representative chromosomes. We'll use red to represent the paternal chromosomes and yellow to represent the maternal. Notice that this cell has diffuse chromatin, and we can't see individual chromosomes yet.

[21] The process of meiosis will involve two divisions: the first meiotic division, or meiosis I, and the second meiotic division, or meiosis II. Look at the entire outline in Section 2 and you'll notice that we use the familiar names of the stages – prophase, metaphase, anaphase and telophase. But make sure you understand that these are two successive divisions, so always refer to exactly which stage you mean. Let's begin.

[22] We'll use this cell to represent interphase I of meiosis I. The chromosomes wouldn't actually be visible as they are here, but just so we can see what's going on, draw in the four replicated chromosomes. Notice the two pairs of homologous chromosomes: a pair of long chromosomes and a pair of short ones. Each strand of DNA has replicated just as it would have in mitosis, so each chromosome is composed of two chromatids held together by a centromere.

[23] Next is prophase I, and this is where the chromosomes would actually become visible. But notice what has happened to our homologous pairs – they have identified the other member of the pair within the cell and are now in close proximity. We'll watch for this significant event called synapsis later in the lab, so complete your drawing of this important stage carefully. Make a note of synapsis on your drawing of Prophase I.

[24] In metaphase I, the attached homologous pairs line up on the equator. Again this should look very different than the metaphase of mitosis. The arrangement of chromosomes at this stage is very important as well, so draw this stage carefully.

[25] In anaphase I, the maternal and paternal members of each homolgous pair move to opposite ends of the cell. Notice these are still full chromosomes because the centromeres have not split as would have in mitosis. In this stage of meiosis, each chromosome is still composed of two chromatids.

[26] In telophase I, the homologous chromosomes have arrived at the poles and cytokinesis begins. The cells will split and complete meiosis I. Notice the two daughter cells at this point – they are already different from each other. What else do you notice at this point? Click on the correct answer.

[27] That's right – these two cells are now haploid. We've got both a long and short chromosome in each cell, so we know we have a full complement of genes in each cell, but what about the configuration of the chromosome? We can't have a viable gamete with the chromosomes still composed of two chromatids. Those will have to be separated from each other with our next division. Make sure your drawing of telophase I is complete before you continue.

[28] These two cells will now undergo the second meiotic division. The good news is that it will look very familiar because is almost exactly like mitosis. We don't need to replicate the DNA here, we just need to divide up those chromatids. Complete your drawing of interphase II, even though most cells spend very little time at that stage, since no additional replication of chromosome material is necessary.

[29] In prophase II, the cells looks very much as we left them in telophase I. The nuclear membrane disappears and new spindle fibers will form.

[30] In metaphase II, each chromosome lines up on the equator of the cell.

[31] In anaphase II, the centromeres split and the daughter chromatids move to the opposite ends of the cell. Each is now called a chromosome.

[32] Telophase II completes meiosis II and let's see what we have. Each cell is haploid and has one of each kind of chromosome... a long one and a short one. Note that there are two kinds of gametes. Two of the four have a long paternal and a short maternal; the other two have a long maternal and a short paternal chromosome. They didn't have to turn out this way though – take a look back at your metaphase I drawing.

[33] What if the chromosomes had lined up this way on the equator during metaphase I? That would have resulted in different looking cells at the end of meiosis II. You don't have to draw this arrangement, but it keep in mind as we continue the lab – you'll thank me later. Okay, now back to our original ending cells.

[34] These cells can now develop into gametes and the work of dividing up the genetic material is complete.

[35] Let's take one more look at mitosis compared to meiosis, this time with chromosomes shown. Very different, isn't it? Your lecture instructor loves asking about the differences between these, so take a careful look at this diagram and continue when you're ready to answer some questions.

[36] Where would each of these processes happen?

[37] Which stage of meiosis is shown here?

[38] Great! Make sure you've answered all the questions at the end of Section 2 and then I'll let you try meiosis with some chromosome models in Section 3. I know! I can hardly stand the excitement myself!

Section 3 – A Model of Meiosis

[39] For this part of the lab, you'll move the chromosome models around your booth to simulate the process of meiosis. You'll have to imagine the margins of the cell, or you can just use one of the white boards in the lab that shows the outlines of the cells, if you wish. Pay attention to the movements of the chromosomes, because in Section 4 of today's lab you'll be asked to demonstrate the entire process to the lab instructor. Sounds fun, huh?!

[40] Find the plastic bag with the chromosome model. At the beginning of Section 3 there is a diagram of what the bead arrangement should be before you continue. *Make sure* your model chromosomes are in this configuration before you begin. See the instructor if you need any extra beads.

[41] Your model organism will have a diploid number of 4 chromosomes, so that should certainly make it easier than trying to manage the 46 chromosomes that a human would have. You will use red and yellow strands to represent paternal and maternal chromosomes just like in your summary drawings.

[42] The chromosomes are inscribed with letters indicating the positions of various genes. For now we are simply interested in the pattern of how these genes are distributed to the new cells. We will get to the meaning of this genetic material in next week's lab, but I wanted to pick a few interesting ones that you've probably heard of such as the genes for what type of blood we have, the color of our eyes and the number of fingers we have.

[43] Complete Section 3 following the instructions in your lab book. Come back when you are ready for some questions.

[44] What is independent assortment? Record the definition when you get the correct answer.

[45] Look at the cells shown here – which of these gametes would be possible if this beginning cell underwent meiosis?

[46] Independent assortment is an important source of variation in gamete formation, so let's look at the significance of this random alignment one more time. Independent assortment shuffles the chromosomes – even though the cells are identical in their genetic makeup when they start, each cell undergoing meiosis will deal out different complements of chromosomes.

[47] If we use a deck of cards to illustrate this concept, the red cards will represent the maternal chromosomes and the black cards represent the paternal chromosomes. This "organism" has a total of 20 chromosomes, and we know that the each gamete this organism produces will only get 10 chromosomes.

[48] But which 10 will be passed on to the gamete? The shuffling of this deck will occur on the metaphase plate – the chromosomes are free to rotate independently of each other allowing many, MANY possibilities. Here are just few examples of possible Metaphase I arrangements.

[49] Now think how this chromosome shuffling would affect a human cell undergoing meiosis – each chromosome has two possible positions on the metaphase plate, and there are 23 pairs of

chromosomes, so that's 2 to the 23rd possibilities or over 8 million different types of gametes produced by a single person.

[50] Remember that every cell starting meiosis in an individual has the same 46 chromosomes, but the process of meiosis insures that all the gametes will be different.

Section 4 – Meiosis Demonstration

[51] Before we look at another source of variation, I want to make sure, *absolutely* sure that you understand how the chromosomes end up in their respective cells. Uh oh, this can't be good...

[52] When you're ready, take your chromosomes up to the white board next to the instructor's table and demonstrate meiosis from replication all the way through the formation of four gametes. To take a little of the stress out, you *may* use your illustrations from Section 2 as a guide, or you *may* do it with a lab partner helping you if you get stuck, but we'll be extra impressed if you can do meiosis all the way through without any help. Come back to the program after you've had your book signed. Good luck!

Section 5 – Crossing Over

[53] Now that you thoroughly understand the movements of the chromosomes during meiosis, we will look at one more source of variation in the production of gametes. This phenomenon is called crossing over. Homologous chromosomes in close proximity will exchange pieces of genetic material - in essence, shuffling the genetic material between the maternal and paternal chromosomes. Record this definition in your lab book.

[54] This phenomenon happens in prophase I of meiosis when the homologous chromosomes synapse and physically join together. The arms of the chromatids will twist around each other, and this is when segments of DNA will actually change places. Here is a photograph with this remarkable event happening.

[55] Crossing-over is not uncommon and doesn't damage the chromosomes in any way since each chromosome carefully exchanges entire genes. Each chromosome can have multiple crossover sites with its homologous chromosome. This will result in varieties of gametes that we wouldn't have predicted simply by looking at the whole chromosomes.

[56] Here is an animation of the process. Notice that the ending chromosomes are the same shape and length as they were when they began, they've just shuffled the information. There would be no way to identify maternal and paternal chromosome material in a pair of real chromosomes just by looking, as genes obviously aren't brightly colored this way.

[57] You can now complete Section 5 by using your model bead chromosomes to see how crossing over works. Come back when you are ready for some questions.

[58] What event in meiosis I shuffled the whole chromosomes?

[59] What event in meiosis I shuffled the genes between chromosomes?

Section 6 – Gamete Variety

[60] For each cell that begins meiosis, many different gamete combinations are possible, even though the cells starting the process are coming from the same individual. Let's look at a couple of examples and keep in mind that meiosis produces variety within the haploid cells that will eventually join up with other haploid cells.

[61] In this first example, a cell happens to have inherited the same genes from their mother and father. Record the genes as shown here. Now what will meiosis do with these homologous chromosomes?

[62] Meiosis will physically split up the homologous chromosomes to separate gametes. Record the cells that result from meiosis. The point of this is that every gamete this individual organism produces will have the same gene for this trait, because they only inherited one kind of gene. Record the final single type of gamete before you continue.

[63] This second example looks similar, but notice now that the genes on the two chromosomes are different this time. Record the position of these genes on the first cell. Meiosis will split the homologous chromosomes and these two genes will end up in different cells. There can only be one gene for this trait per cell, because gametes are haploid. Record the two *types* of gametes produced in this situation.

[64] But that was just looking at one trait. There are thousands of genes and multiple chromosomes to arrange. In this third more complicated example, we're going to keep track of two genes on two different pairs of chromosomes. But notice that again, this organism just happened to inherit identical genes from both parents, so what sort of gametes would be produced here? Based on what you know about meiosis producing complete sets of chromosomes, take a guess!

[65] If you realized that each gamete would have the same combination of single genes, I'm impressed! Record the arrangements of genes in this example and look ahead to the resulting gamete – all the gametes that this individual produces will have the same information because the genes on the two pairs of homologous chromosomes were exactly the same for these particular traits. No matter how these genes are shuffled, the result is the same. Record the single gamete that is possible before you continue.

[66] For our fourth and last example, still looking at two traits, you can see that there will be more combinations possible. For this one you'll have to remember that independent assortment and crossing over will still shuffle all the genes, but the gamete can still only contain one of each. Try to predict the gametes in this situation.

[67] Did you get four unique combinations while still making sure each gamete got a full set of information? Excellent! Isn't it amazing that this careful orchestration of genes is happening every day, every where for every sexually reproducing organism on earth?

[68] Now take a minute and look over all the gene combinations you created in the gametes of Section 3 and 5. Those were results looking at just three genes for eye color, blood type and the numbers of fingers. I think by now you understand the immense variety of gametes possible.

[69] So based on these very simple patterns and remembering all the combinations that are possible, how many types of gametes could *you* produce? We would have to keep track of your 46 chromosomes and tens of thousands of genes - it's too much to think about right now, isn't it? But for now, read over the summary at the end of Section 6 and make sure you understand the mechanism behind each statement.

Section 7 - Meiosis in Humans

[70] You now have a good idea about where, why and how meiosis happens in general, but now let's look specifically at humans. To do this, we'll have to take a peek into the gonads. Ready? Here we go...

[71] In human males, sperm is produced in the seminiferous tubules of the testes. This specific type of meiosis is called spermatogenesis, which translates as "sperm making". Here you can see a cross-section of this very specialized tissue. Remember, this is a type of cell division found nowhere else in the human body.

[72] Meiosis in human males takes about 3 days. From the seminiferous tubules, the sperm travel to the epididymis where it will take a few more weeks before these cells are mature and fully functional as sperm.

[73] Spermatogenesis usually begins at about 12 years of age and continues for the rest of a man's life. Several hundred million sperm cells are produced daily by healthy young adult males, and between 200 and 600 million sperm cells are normally released in each ejaculation.

[74] Gamete production in males is rather straight-forward – one diploid cell produces four haploid sperm as shown in the diagram at the beginning of Section 7. In human females, however, it's quite a bit more complicated both in the structures and the timing. Egg production is called oogenesis and it happens only in the ovaries. But do you see something different in the summary diagram of oogenesis compared to spermatogenesis? Take a look at telophase I.

[75] Did you notice how uneven the cytokinesis was in telophase I? Both cells will have a haploid number of chromosomes, but only one cell garnered all the cytoplasm. Look ahead to telophase II and you'll see the same unequal cytokinesis. There is an important reason for this - the resulting egg has a much better chance at survival, because it is the egg which supplies the embryo with nutrients until it implants on the uterus several days after fertilization. Answer the question about why the cytokinesis is different before we go on.

[76] Look closely at the end of telophase II. Notice the little leftover cells from the unequal cytokinesis? These cells are called polar bodies and they would never be able to function as gametes – they have no cytoplasm. So is the single egg that is produced still a haploid cell? Yes, and it's because of those polar bodies. These leftover cells contain chromosomes from the meiotic division, but no cytoplasm, so the number of chromosomes in the egg is correct.

[77] What about all the variation we learned about between gametes? Well the variation is still possible, because the variation we refer to is based on the lifetime production of eggs, so a human female would have the same huge variety of eggs possible due to independent assortment and

crossing over, just like a male. It's just that the female doesn't produce as many gametes at a time as a male does.

[78] The timing of meiosis in human females is quite complicated as well. Before a girl is born, her future eggs in her ovaries begin meiosis, but stop after prophase I. They remain in this suspended state of division until puberty, when hormones will signal the continuation of meiosis.

[79] Each month, a single egg is released into the fallopian tubes at ovulation. But the other strange thing is that this "egg" has not actually finished the final division of meiosis – that won't happen until a sperm penetrates the egg and signals that it's time to finish up meiosis and the true final egg and last polar body are produced.

[80] Here you can see a photograph of a human egg at the moment of ovulation. Like all eggs, it is a relatively large cell – much larger than most of the other cells of the body and much larger than sperm, which must be quick and nimble swimmers to reach the egg.

[81] So human egg and sperm production is quite different. Look over the two summary diagrams for meiosis in males and females and click on the answer to this question.

Section 8 – After Meiosis – Fertilization

[82] Regardless of how gamete formation may differ from sex to sex or from species to species, all gametes have one role: to participate in fertilization and produce the next generation.

[83] Remember at the beginning of the lab when I said that the two processes of mitosis and meiosis were parts of a larger cycle? Well we've covered both topics thoroughly enough by now that I don't think you'll have too much trouble filling in this summary diagram. Drag the labels to the correct parts of the diagram and fill them in on your lab book when you have them all correct.

[84] Let's look at an organism with a very simple strategy for reproduction, the sea urchin. Sea urchins don't even have to be near each other to have sex – the male and female simply release their gametes out into the open ocean and it's all a matter of chance whether or not the eggs get fertilized. To make this successful, both the male and female must produce thousands of gametes and do it with rather precise timing.

[85] This photo shows the sea urchin sperm swarming around the as yet unfertilized egg. Remember that all of these cells are haploid. Now what would happen if two or more sperm entered the egg? That would be genetically disastrous.

[86] To prevent extra sperm from entering an egg, a protective membrane lifts off the surface of the egg as soon as soon as the first sperm enters. This special structure is called the fertilization membrane and keeps any other sperm from penetrating the egg. Sketch the appearance of these cells, the unfertilized sea urchin egg and the zygote, in your lab book. Answer the next few questions about sea urchins before you continue.

[87] The sea urchin system of reproduction may seem a little haphazard, but urchins don't have to spend any time or energy raising the kids. Once the gametes are released, mom and dad urchin are done.

[88] The reproductive strategy for humans is quite different for obvious reasons – human females can get away with producing just one egg a month because the egg could only be in one place – inside the female. With internal fertilization found in mammals and a lot of other vertebrates, you don't need thousands of eggs ready at one time.

[89] Once a human egg has been fertilized, the nuclei of sperm and egg fuse and the zygote will begin to divide. A few days later, the embryo will implant on the uterus and obtain all the nutrients it requires for growth from the mother. Notice that we've arrived back at the process of mitosis – growth of the embryo. The cycle has begun again.

Section 9 – Early Development

[90] We will continue our discussion of development with an organism closely related to the sea urchin, starfish. Find the microscope slide with starfish development in your booth and start with scanning. This slide will contain a variety of developmental stages mixed together, so let me show you what to look for as we go. And please be very careful with these slides and don't use high power!

[91] Soon after fertilization, the zygote will undergo cleavage to form two cells. Since this is a mitotic division, each cell will have a nucleus with genetic information identical to that of the zygote, but will be half the size of the zygote. These cells don't have time to grow. Find a two-cell stage in your slide and sketch it in the first box of Section 9.

[92] The cells of the 2 cell stage will divide again by mitosis to form four cells and again the cells are reduced in size. Find the actual four cell stage on your microscope slide and sketch it in your lab book.

[93] The four-celled stage is followed by an eight-celled stage, then 16, 32, and so on. When the embryo reaches these stages, it becomes difficult to count the number of cells, and the overall appearance of the embryo is called a morula, which translates as "berry". The image shown here is an illustration of a morula, so look for this basic shape to find an actual starfish morula in your microscope slide and sketch it in your lab book.

[94] As cell division continues, the cells begin to organize into an outer layer with a hollow center. This hollow ball is called a blastula. Find a blastula to sketch in your microscope slide before you go on. Make sure you have found a blastula and not just a zygote – look for small cells on the outer layer.

[95] The embryo now begins to develop a mouth – one side of the blastula starts to fold in, in a process called invagination. Once you see this invagination, our embryo is called a gastrula and the formation and differentiation of tissues has begun. A gastrula from a sea cucumber is shown here – your starfish gastrula will be very similar.

[96] As development and differentiation continue, a free swimming larval stage called a pluteus larva forms. Since this stage will probably not be in your microscope slide, we have placed a pluteus larva stage on a demonstration scope in the lab, or you can draw this one on the screen. At this stage the larva feeds and grows, but notice it doesn't look anything like a starfish at this stage. This is true of many organisms – the embryonic stage can be dramatically different than the adult organism. Make sure you have found and sketched all the starfish stages before you continue.

[97] Let's finish up today by making sure you appreciate the significance of the process of meiosis. All sexually reproducing organisms have an incredible genetic variety. When a sperm fertilizes an egg, the genes of each combine in a way that has probably never existed before.

[98] Remember the 8 million kinds of gametes that every human can potentially produce even without involving crossing over? Well that unique gamete is combining with another person's unique gamete. This unique genetic identity doesn't just apply to how a person looks; it also determines everything about their physiology. Can they tolerate lactose? Are they allergic to certain substances? What type of blood do they have? Will they develop arthritis early or late in life?

[99] The results of these unique combinations will be the subject of next week's lab - patterns of genetic inheritance. See you next week!