Embryo Clay Model Embryogenesis and Stem Cell Development

Objective:

- To identify stages and locations of early embryonic development
- To see at what point stem cells are totipotent, pluripotent, and multipotent at various stages of development
- To understand the origin of cells used to create stem cell lines.
- To understand that all tissues of the body are derived from three populations of cells (ectoderm, endoderm and mesoderm)
- To provide a hands on activity to better visualize the embryo in 3D.

Introduction:

In this activity students will make clay models of embryos at various early stages of development while learning important terminology, and the potency of stem cells at various stages. Students will then use the models they constructed to map the journey of the developing embryo along the uterine tube/oviduct toward the uterine cavity.

Materials:

- 4 colors of modeling clay or play-doh (5oz of one color and 1oz of 3 other colors) per station or group.
- Paper plates for a work surface
- 6 (2x2 inch) parafilm strips per group or station. Plastic wrap can also be used.
- 1 pair of scissors per group or station
- Modeling tools (plastic knives, cap of a pen)
- Embryo Clay Model Table/Worksheet

Procedures:

PART I - The "Embryo Clay Model Table/Worksheet" is a visual and instructional guide for constructing the clay embryos. Teachers can demonstrate each step while explaining the structures if desired. Students can work individually or in groups.

1. START by reading the instructions below.

- Select a 5 oz. container/color of clay
- Use this clay to make 5 ping pong size balls and set aside
- Read the description for the "DAY 1 zygote, examine the figure, and follow the directions for construction.
- After completing construction of the day 1 zygote continue on down the worksheet.

A glossary of terms that have been bolded follows PART II can be used to fill in answer The questions at the end of this activity.

Embryo Clay Model Table/Worksheet		
STAGE/DESCRIPTION	FIGURE	DIRECTIONS
 Day 1 Zygote Fertilization begins when a sperm penetrates through the zona pellucida (ZP - membrane of the oocyte) Fusion of the oocyte and sperm nuclei (pronuclei) marks the formation of the zygote (2n). Embryonic stem cells are totipotent at this stage. 		 Select one of the 5 balls you made earlier and wrap it in parafilm. The parafilm represents the ZP surrounding the zygote.
 Day 2 2 Cell Zygote Zygote goes through 1st division within the ZP producing 2 daughter cells (blastomeres) Embryonic stem cells are totipotent at this stage. 		 Select another ball and divide it into 2 equal size spheres (blastomeres). Wrap this 2 cell zygote with parafilm (ZP).
 Day 3 – 4 4 Cell Zygote and 8 Cell Zygote 2 blastomeres divide forming 4 then 8 blastomeres with each subsequent division each blastomere decreases in size as all the cells are contained with in the ZP. Embryonic stem cells are totipotent at this stage. 	1 - ZP $2 - Blastomere$ Dashed lines indicate cross-sectional cut.	 Select another ball and divide it into 4 equal size spheres (blastomeres). Gently press them into a sphere Wrap with parafilm (ZP). Repeat this procedure forming an 8 cell zygote Wrap with parafilm.

STAGE/DESCRIPTION	FIGURE	DIRECTIONS
 Day 4 Morula Early embryo stage which consists of about 16-32 cells is now called a morula. After reaching the 16 cell stage the cells of the morula begin to differentiate. Embryonic stem cells are <i>pluripotent</i> at this stage. 		 To make a 16 cell morula again select a sphere and divide it into 16 equal size pieces, role each into a ball and gently press them into a ball. Wrap the morula with parafilm. Note: As the embryo/zygote continues to develop it remains the same size/volume.
 Day 5 Blastula Together a spherical layer of about 120 cells (trophoblast) surrounding a fluid filled cavity (blasocoel) forms the blastula. Cell differentiate into the inner cell mass (ICM) or epiblast and the outer trophoblast. Trophoblast cells secrete and enzyme that erodes the lining of the uterus (important for implantation) Embryonic stem cells are pluripotent at this stage. 	 I - ICM 2 - ZP 3 - Trophoblast 4 - Blastocoel 	 To form the blastula, select a new color of clay and form a ping pong sized sphere. Flatten and shape into a hollow bowl Use pen cap to press cell shape pattern into the "trophoblast". You have just constructed a cross-section model of the blastula. Make a ball about ½ the size of a ping pong ball using either the same or new color of clay. Shape ball into a tube about the size of a pen. Use your plastic knife to cut pea size pieces and shape into balls. Place into the trophoblast to represent cells of the ICM. Wrap blastula in parafilm.

STAGE/DESCRIPTION

Day 13 Gastrula

- During gastrulation, cellular reorganization forms a multi-layered sphere. Many of the cells at or near the surface of the embryo move to a new, more interior location.
- During gastulation, primary germ layers (endoderm, mesoderm, and ectoderm) are formed and organized in specific locations.
- Endoderm, most internal layer, forms the gut and other internal organs.
- Ectoderm, most exterior germ layer, forms skin, brain, and the nervous system.
- **Mesoderm**, middle germ layer, forms muscle, skeletal and circulatory systems.
- Embryonic stem cells are multipotent at this stage.

FIGURE



DIRECTIONS

Between the blastula and gastrula stage the ZP is lost during "hatching."

- To form the gastrula stage mold a new trophoblast as you did above.
- Use 3 colors of clay to make 3 sets of pea sized cells (one set should be the same color as the previous set).
- Arrange the cells into 3 layers, representing the 3 germ layers (endoderm, ectoderm and mesoderm).
- Place cells inside the trophoblast
- The original color can represent the ectoderm
- The remaining colors layers can be arranged to represent the mesoderm and endoderm.
- Do not wrap in parafilm.

PART II – Journey of the Embryo

- 1. Examine the figure on the following page and match the embryo models you constructed with the images on figure.
- 2. Physically place the models on the figure.
- 3. Notice that fertilization occurs in the uterine tube/oviduct near the ovary. The timing of embryo development with it's arrival at the uterine cavity is critical for successful implantation.

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Stages of Development-Early Embryo



Glossary

Blastocoel – The cavity in the blastula of the developing embryo.

Blastocyst – A pre-implantation embryo of 30-150 cells. Consists of a sphere made up of an outer layer of cells (the trophoderm), a fluid-filled cavity (the blastocoels), and a cluster of cells on the interior (inner cell mass).

Blastula – An early Stage in the development of an ovum consisting of a hollow sphere of cells enclosing a cavity called the blastocoel.

Differentiation – the process whereby an unspecialized early embryonic cell acquires the features of a specialized cell such as a heart, liver, or muscle.

Ectoderm – The upper outermost of the three primitive germ layers of the embryo; it gives rise to skin, nerves, and brain.

Embryo – In humans, the developing organism, from the time of fertilization until the end of the eighth week of gestation, when it becomes known as a fetus.

Embryonic stem cells – Primitive undifferentiated cells from the embryo that have the potential to become a wide variety of specialized cell types.

Endoderm – Lower layer of a group of cells derived from the inner cell mass of the blastocyst; it later becomes the lungs, and digestive organs.

Epiblast – Gives rise to the ectoderm and mesoderm. The mesoderm then displaces the hypoblast cells and forms the endodermis cell layer on its inner surface.

Fertilization – the process where by male and female gametes unite.

Gastrula – Animal embryo at an early stage of development in which cells are enclosed in a sheath to form the beginning of a gut cavity.

Hypoblast – The inner cell layer, or endoderm, which develops during the formation of the embryonic germ layers.

Inner cell mass – The cluster of cells inside the blastocyst. These cells give rise to the embryonic disk of the later embryo and ultimately, the fetus.

Mesoderm – The middle layer of the embryonic disk which consists of a group of cells derived from the inner cell mass of the blastocyst. This middle germ layer is known as gastrulating and is the precursor to bone, muscle and connective tissue.

Morula – A solid mass of cells that resembles a mulberry and results from the cleavage of a fertilized oocyte.

Multipotent stem cells – stem cells hat have the capability of developing cells of multiple germ layers. These cells are not able to develop into as many cell types as totipotent or pluripotent cells.

Placenta – the oval or discoid spongy structure in the uterus from which the fetus derives its nourishment and oxygen.

Pluripotent stem cells – A single stem cell that has the capability of developing cells of all germ layers (endoderm, ectoderm and mesoderm).

Stem cell – A cell that has the ability to divide for indefinite periods in culture and give rise to specialized cells.

Totipotent – Having unlimited capability. The totipotent cells of the very early embryo have the capacity to differentiate into extra embryonic membranes and tissues, the embryo, and all postembryonic tissues and organs.

Trophoderm – The outer layer of the developing blastocyst that will ultimately form the embryonic side of the placenta.

Trophoblast – the extra embryonic tissue responsible for negotiating implantation, developing into the placenta, and controlling the exchange of oxygen and metabolites between mothers and embryo.

Yolk sac – Vital to the formation of primordial and other cells that form the embryo. In mammals, it is small and devoid of yolk.

Zona pellucida – A thick, transparent non-cellular layer that surrounds and protects the oocyte.

Zygote – A cell formed by the union of male and female germ cells (sperm and egg, respectively).

Embryo Clay Model Questions

1. Circle the stages at which the embryonic stem cells are totipotent. Underline the stages at which cells are pluripotent. Do nothing below if the stage is multipotent.

Day 1 ZYGOTE – 8 CELL STAGE ZYGOTE – MORULA – BLASTULA – GASTRULA

2. At what stage of development do stem cells first differentiate?

3. At what stage of embryonic development are stem cells used to generate embryonic stem cell lines. Which type of stem cells is found at this stage?

- 4. What types of tissues are formed from the following primary germ layers?
 - a. Endoderm
 - b. Ectoderm
 - c. Mesoderm
- 5. If an embryo entered the uterus at the morula stage would it implant successfully into the uterine wall? Explain you answer.