Totipotency

A human egg can be fertilized by a human sperm either naturally by sexual intercourse or artificially in a laboratory (known as **in vitro fertilization** or **IVF**). A fertilized egg is called a zygote. This cell is **totipotent**. That is, it has the ability to divide and specialize into all the cell types found in a human being and into all of the cell types that form the extra-embryonic tissues, such as the placenta, umbilical cord and amniotic sac. In other words, a zygote has the potential to develop into, and support the development of, a newborn infant if allowed to implant into a functioning uterus. The process by which a zygote develops into an embryo, a fetus and then an infant is, essentially, a process of cell division and increasing specialization of different groups of dividing cells. An infant is composed of trillions of cells, most of which are highly specialized cells in muscles, nerves, the liver, the brain, and so on. The single cell that constitutes the initial zygote is completely unspecialized but has the potential to specialize.

Please note: definitions of terms that are in boldface in the text as well as other terms can be found in the Glossary.

The fertilized egg divides into two cells. These two cells are still unspecialized and therefore are still totipotent. We know this because occasionally identical twins develop in the uterus when these two cells separate completely, divide to produce their own separate supporting tissues (placenta, umbilical cord, and so on), and eventually develop into two infants. Usually, however, the two-celled zygote remains intact and divides into 4 cells. We know that those 4 cells are still totipotent because, by the same mechanism just described for twins, the 4 cells can very rarely separate and develop into 4 individual infants.

Pre-implantation Genetic Diagnosis (PGD)

If the 4-celled zygote divides again into 8 cells, these cells are usually, if not always, still totipotent because identical octuplets have very rarely occurred. These 8-celled zygotes have also been used for a process known as **pre-implantation genetic diagnosis** (**PGD**). In fertility clinics, infertile couples can use IVF to create a zygote. Eggs from a woman are combined with sperm from a man to create zygotes. These zygotes then begin to divide to become embryos in a Petri dish in a lab. Sometimes, the couple asks to have genetic testing conducted on the embryos. PGD can be performed by carefully removing one of the cells at the 8-cell stage. The cell is then examined for genetic abnormalities. The remaining 7-celled zygote is allowed to continue to divide, usually resulting in what appears to be a perfectly normal infant. However, it also means that the single removed cell could also have the potential to form an identical twin of the

remaining 7-cells zygote, presenting an ethical problem for some who consider such a totipotent cell to be a potential human being.

Harvesting Embryonic Stem Cells for Research Purposes

When the embryo consists of about 150 cells, known at this stage as the **blastocyst**, the outer cells have differentiated (specialized) into cells that will only develop into supporting cells such as the umbilical cord or placenta. The remaining inner cells are now considered **pluripotent** rather than totipotent. They are still capable of differentiating into a wide variety of cells under the proper environmental and genetic influences. However, if separated from the outer cells, they cannot form a human infant if placed in the uterus and can no longer differentiate into the umbilical cord or placenta.

It is these inner cells that are so sought after by scientists who do research with human embryonic stem cells. The main source of such cells is extra embryos that are produced during IVF and are no longer wanted for producing human infants. Scientists separate out this inner cell mass from the outer cell layer, destroying the embryo in the process. These inner cells are then cultured in laboratory dishes as embryonic stem cells, which are used in experiments. Such destruction of human embryos raises important ethical problems for many people. Some feel that embryos are not fully human beings, using that claim to justify killing them for research to develop therapies to help others later. Others feel that the embryo has the status of a human being and that killing the embryo is immoral.

Alternative Sources of Human Embryonic Stem Cells

Somatic Cell Nuclear Transfer (SCNT)

In recent years scientists have tried to develop other sources of cells that have similar physical, physiological, and genetic characteristics to those of embryonic stem cells One of the first such sources involved making an embryo in the laboratory by a method called **somatic cell nuclear transfer** *(*or **SCNT***),* the same method by which animals such as Dolly the sheep have been cloned (some call the cell cluster produced by this method an **embryoid** rather than **embryo** to distinguish it from an embryo produced from the merger of a sperm and an egg. Some consider the use of embryoids to be less morally objectionable than the destruction of embryos as a source of stem cells for research). In this technique, a nucleus from an ordinary cell, like a skin cell, from a particular animal is joined with an egg cell from that same species whose nucleus had previously been removed. The new cell is stimulated, usually by electrical current, causing it to divide and develop into cells that look like, and function like, embryonic

stem cells produced by joining a sperm cell and an egg cell. Although this process has been successful in mammals like Dolly the sheep, it has not yet succeeded in humans. If it did succeed in humans, the result would be an embryo from which stem cells could be obtained. In this case, the stem cells would be genetically identical to the person who donated the nucleus for the somatic cell nuclear transfer (There is one exception: you'll recall from the previous discussion in Genetics 101, A Sidebar: DNA Outside of the Nucleus, that a very small amount of DNA is located not in the nucleus but rather is in the mitochondria. This DNA would be derived from the donated egg).

Induced-Pluripotent Stem Cells

More recently, scientists have discovered newer ways to construct cells in the laboratory that also have many characteristics similar to those of embryos. Instead of being created by the union of a sperm and an egg or by somatic cell nuclear transfer as described above, these cells are produced from an ordinary body cell like a skin cell, and not just its nucleus. These ordinary somatic cells are manipulated in the laboratory to lose their normal functions while taking on characteristics similar to those found in pluripotent stem cells. These now undifferentiated (unspecialized) cells can then be "induced" (that is, directed under special laboratory conditions) to re-differentiate (re-specialize) into a cell with a new, desired function that may be completely different from its original function. For example, a skin cell might be de-specialized, then re-specialized to function like a cell that now produces insulin, like certain cells normally found in the pancreas. This cell could then be replicated (multiplied) in the laboratory and could be transplanted into a person with diabetes whose own insulin-producing cells no longer work.

These **induced-pluripotent stem (iPS) cells** are considered by some to be more ethically acceptable for developing new therapies. For at least two important reasons, some consider this method more ethically acceptable than using leftover embryos after IVF or using SCNT: 1) these cells are not produced involving the use of natural embryos, produced from sperm or eggs that must be destroyed to harvest the stem cells and 2) this method does not require a supply of human eggs that would involve ethical problems such as risky drug therapy to produce the eggs from female donors and tempting women in need of money to take those risks and accepting payment for donating their eggs.

Human Engineering and Cloning

It has been proposed that combining stem cells and embryo cloning could allow **germline genetic engineering**. In this process, pluripotent stem cells would be removed from an embryo or embryoid (that is, stem cells produced by SCNT from iPS cells

described above). These cells would then be infected with a virus that contains whatever genes someone desired to transfer into the cells. The cells could be tested for successful incorporation of the desired gene or genes into the nucleus of those cells. Cells that have been successfully engineered could have their nuclei removed and transferred to denucleated eggs (recall that this is the somatic cell nuclear transfer technique described in Transgenics, Animal Cloning). The resulting cell could be allowed to develop into an embryo, then implanted into the uterus of a surrogate mother. If a fetus developed fully and was born, that newborn would carry the engineered genes in every one of his or her cells. If that newborn grew to adulthood and reproduced, his or her offspring would also possess these introduced genes. It is important to note that this process has not at this point been achieved in primates, including humans, but scientists are currently working to develop this process.