

National Institutes of Health (NIH) Update on Existing Human Embryonic Stem Cells

August 27, 2001

On August 9th, President Bush announced that federal funds may be awarded for research using human embryonic stem cell lines that meet certain criteria. Such research is now eligible for federal funding as long as the derivation process (which begins with the destruction of the embryo) was initiated prior to 9:00 p.m. EDT on August 9, 2001. These stem cells must have been derived from an embryo that was created for reproductive purposes and was no longer needed. In addition, informed consent must have been obtained for the donation of the embryo and that donation must not have involved financial inducements.

The NIH is expeditiously implementing the President's new policy. It is our hope that federally-funded investigators will take full advantage of this new opportunity to conduct research on existing human embryonic stem cells and explore the enormous promise of these unique cells, including their potential to produce breakthrough therapies and cures. With the help of the scientific community, this research will mark the beginning of a new era in modern medicine.

As discussed below, investigators from ten laboratories in the United States, Australia, India, Israel, and Sweden reported to the NIH that they have derived stem cells from 64 individual, genetically diverse blastocysts. These derivations meet the President's criteria for use in federally-funded human embryonic stem cell research. The NIH is working to ensure that the scientific community will soon be able to use federal funds to tap the extraordinary research potential of human embryonic stem cells. The NIH has met or spoken extensively with each of the investigators who have derived these cells. These scientists are very interested in working with the NIH and the research community to establish a research infrastructure to ensure both the successful handling and the use of these cells in the laboratory.

What do we know about existing human embryonic stem cell derivations?

At the request of the Secretary of Health and Human Services, Tommy G. Thompson, the NIH has identified investigators from ten laboratories in the United States and around the world who reported that they had derived human embryonic stem cells from 64 individual, genetically diverse blastocysts. All of the existing cells reported to the NIH meet the President's criteria. The scientists who developed these stem cell lines report that the cells are viable, show characteristic stem cell morphology, can be maintained frozen, as well as in culture, and have undergone at least several population doublings. The majority of these cells were reported to express all of the markers known to be associated with human embryonic stem cells, including stage specific embryonic antigens (SSEA-3 and SSEA-4), the enzyme, alkaline phosphatase, and tumor rejection antigen 1 (TRA-1-60 and TRA-1-81). The scientists reported to the NIH that the cells could be frozen and thawed and continue to grow while maintaining their karyotype. They also reported that in many cases they had assayed the cells for pluripotency by injecting the cells into immune-deficient mice and showing the formation of ectodermal, endodermal, and mesodermal tissues. Further information about these human embryonic stem cells will be made available to the scientific community at the discretion of the investigators who derived them.

The following entities reported to the NIH that they have derived human embryonic stem cells that meet the President's criteria:

Name	Number of existing stem cell lines reported to NIH
BresaGen, Inc., Athens, Georgia	4
CyThera, Inc., San Diego, California	9
Göteborg University, Göteborg, Sweden	19
Karolinska Institute, Stockholm, Sweden	5
Monash University, Melbourne, Australia	6
National Center for Biological Sciences, Bangalore, India	3
Reliance Life Sciences, Mumbai, India	7
Technion-Israel Institute of Technology, Haifa, Israel	4
University of California, San Francisco, California	2
Wisconsin Alumni Research Foundation, Madison, Wisconsin	5

What do we know about the derivation and characterization of human embryonic stem cells?

The NIH recently issued a report titled, *Stem Cells: Scientific Progress and Future Research Directions*. Chapter 3 and Appendix C of this report describes the process by which human embryonic stem cells are derived, including the various methods for removing the cells from the blastocyst and the ways in which they are cultured and grown.

Because human embryonic stem cell research is in its initial stages, there is to date no universally accepted standard for determining what characteristics will predict the ability of such cells to be, for example, differentiated or, ultimately, useful for the development of therapies. Currently, scientists seeking to characterize embryonic stem cells describe the morphology of the cells and the detection of specific cell surface markers, such as stage specific embryonic antigens (SSEA-3 and SSEA-4) and the enzyme, alkaline phosphatase, as essential characteristics of embryonic stem cells. They also require evidence that the cells are pluripotent by injecting them into immuno-deficient mice to see if they generate differentiated cells from all three germ layers. Some scientists require evidence that a putative human embryonic stem cell line maintains its pluripotency for 12 months or more, while sustaining a normal karyotype. Still other investigators look for evidence of additional cell surface markers, including tumor rejection antigen 1 (TRA-1-60 and TRA-1-81), expression of certain genes such as the transcription factor, Oct-4, and/or high levels of telomerase activity. In this regard, it is noteworthy that there have been no reported comparative studies on the characteristics of human embryonic stem cells from different derivations.

How is the NIH implementing the President's decision?

In order to facilitate research using human embryonic stem cells, the NIH is creating a Human Embryonic Stem Cell Registry that will list the human embryonic stem cells that meet the eligibility criteria. Specifically, the laboratories or companies that derived the cells listed on the Registry will have provided a signed assurance that the derivation process was initiated prior to 9:00 p.m. EDT on August 9, 2001, informed consent was obtained for donation of the embryo, the cells were derived from an

excess embryo that was created for reproductive purposes, and there were no financial inducements for the donation of the embryo for research. The Registry will be accessible to investigators on the NIH Home Page (www.nih.gov).

Initially, the Registry will contain basic information about the cells. This information will include a unique identifier; the name of the company or laboratory that derived the cells; contact information for the company/laboratory; and an assurance that the cells meet the President's criteria. In the future, to further assist researchers, additional information may also be included in the Registry, such as details about the derivation of the cells, the number of passages, culture conditions, and growth characteristics; a description of efforts to characterize the cells, including molecular markers and evidence of pluripotency; relevant publications; DNA fingerprinting data; and quality assurance data, such as the results of tests for Mycoplasma species and standard human pathogens.

The NIH is working expeditiously to ensure that the Registry will be operational as soon as possible. General questions or comments about the Registry should be addressed to stemcellregistry@od.nih.gov. Researchers who are interested in studying or using particular human embryonic stem cells in their research will be expected to contact the company/laboratory directly to arrange for access to these cells. Investigators should be aware that, in some cases, laboratories that have derived the cells may need to expand to reach larger numbers for the purposes distribution, and that, in other cases, the derivations are still in the early stages of characterization and, thus, may not be immediately available.

With regard to the funding of research on both embryonic stem cells and adult stem cells in humans and animals, the NIH welcomes investigator-initiated grant applications proposing research using such stem cells, including requests to use existing funds or for supplements to existing grants to conduct such research. The NIH is also exploring a number of initiatives to facilitate research on all forms of stem cells. To hasten the development of a program of research and to stimulate submission of grant applications in this arena, some Institutes and Centers will be issuing Program Announcements to describe new, continuing, or expanded interests relevant to stem cell research, such as new approaches to the characterization of stem cells or the development of methods to differentiate cells into specific somatic cells for study. Other Institutes and Centers may issue specific Requests for Applications, which invite grant applications, include a special receipt date, and a set-aside of funds. Another possibility may be provision of resources (using contract or other mechanisms), in order to ensure adequate production of cells, means for their distribution, and adequate training of researchers as to how to maintain the cells.

What are the technology transfer issues relevant to NIH-funded investigators gaining access to human embryonic stem cells?

As with other biomaterials that are a necessary prerequisite for the conduct of NIH-funded research, NIH funding recipients will be responsible for arranging access to particular cells that they determine are necessary for their research. The NIH is interested both in accessing cells for use in its intramural research program, as well as in facilitating access for the broader research community. The NIH's goal is to facilitate the transfer of cells from providers under acceptable conditions and with as little administrative burden as possible.

Recently, the NIH has been meeting with potential cell providers to discuss these topics. In these meetings, the NIH has, on behalf of its intramural investigators, initiated negotiations with organizations that have derived human embryonic stem cells. Although the NIH does not have the authority to negotiate agreements on behalf of grantee institutions or third parties, it has been the NIH's experience

in other cases that the agreement into which it enters may serve as a blueprint for separate agreements by NIH-funded investigators, should their institutions choose to use it.

Some scientists have asked about the effect that patents that have been filed and/or issued over the past few years will have on human embryonic stem cell research. The NIH wishes to emphasize that the issuance of patents on new discoveries need not adversely affect continuing research, provided that the patent owners devise a licensing and sharing strategy to allow basic research to proceed. Experience has shown that conditions imposed by patent owners can be crafted both to ensure research uses and to provide appropriate incentives for commercial development.

In the United States, a patent on human pluripotent embryonic stem cells was issued to the Wisconsin Alumni Research Foundation (WARF), of Madison, Wisconsin. Pursuant to its rights under its patents, WARF negotiated a commercial license, for a limited number of cell types, to Geron Corporation of Menlo Park, California. WARF, as well as many other cell providers, have publicly stated that they are very interested in making their cells available for use in federally-funded research. Although the specific terms and conditions of availability must be determined between providers of the cells and the recipients, the NIH is pleased by the willingness of the researchers who have derived cells to make them available for use by federally-funded researchers. The NIH urges all providers to make their cells available in accordance with its policy on access to research tools, "Sharing of Biomedical Research Resources, Principles and Guidelines for Recipients of NIH Research Grants and Contracts," which is available at:

http://ott.od.nih.gov/NewPages/RTguide_final.html

What happens next?

Some scientists have questioned whether limiting federally-funded research to stem cells derived from existing embryos will inhibit NIH-funded investigators from conducting research on stem cells that represent, for example, all of the major histocompatibility complex antigens or on cells of sufficient genetic diversity. Others have suggested that these existing cells will not meet future demands for clinical trials of potential cell-based therapies. As with any new technology, the characteristics and utility of any of the existing stem cell derivations will only fully emerge with considerable future research. The NIH believes that much basic research can and should be conducted using existing stem cells before any conclusions can be reached regarding the therapeutic potential of these unique cells.

For example, scientists will seek to determine the best conditions for growing the cells and directing their differentiation into specialized cells, such as neurons, muscle cells, and insulin-producing cells. Investigators will also need to learn about some of the key genes that control the capability of an embryonic stem cell to proliferate in an undifferentiated state. This will have important consequences for all types of stem cells, embryonic, fetal, and adult. Scientists will also need to conduct basic research to identify and try to isolate to purity each cellular intermediate between human embryonic stem cells and a mature cell type, such as a cardiac muscle cell or a neuron. Information from the sequencing of the human genome will subsequently need to be used to determine which genes are turned on or off at each stage of differentiation to further enable the development of specific cell-based therapies. Other researchers may choose to facilitate the application of stem cells in proof-of-concept studies using animal models and in culture systems that will determine how such cells evoke their biological effects in disease. Research on some of these existing cells will enable a broad range of safety testing in culture and in animal model systems.

Research using human embryonic stem cells will also facilitate the development of approaches to avoid immune rejection transplanted cells. It will also enable researchers to explore the expression of proteins in stem cells as they become specialized. Scientists will also begin to explore the functional aspects of newly differentiated cells, such as the expression of hormones and growth factors and the conduction or contraction characteristics of nerve, heart, or skeletal muscle, as well as their integration into and ability to survive in target tissue.

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The NIH is pleased to be implementing the President's new policy regarding federal funding of stem cell research. We will do all we can to ensure that the scientific community has an opportunity to fully and swiftly investigate the promise of human embryonic stem cell research. We urge federally-funded researchers to begin their explorations with the profound hope that we stand at the threshold of a true breakthrough in our ability to treat disease and disability.