“Alternative Sources of Human Pluripotent Stem Cells”

Human pluripotent stem cells are valued for their potential to form numerous specialized cells and for their longevity. In the US, where a portion of the population is opposed to destruction of human embryos to obtain stem cells, what avenues are open to scientists for obtaining pluripotent cells that do not offend the moral sensibilities of a significant number of citizens? It is this question that the official position paper, or white paper, “Alternative Sources of Human Pluripotent Stem Cells,” published in May 2005 by the President’s Council on Bioethics under the chairmanship of Leon Kass, seeks to answer. Three experts external to the council, Andrew Fire from the Stanford University School of Medicine, Markus Grompe of the Oregon Health and Science University, and Janet Rossant from the Samuel Lunenfeld Research Institute in Toronto, also reviewed the white paper prior to publication.

In the paper, the Council provides a brief overview of four proposed methods for obtaining human pluripotent stem cells without destroying human embryos. In the first method, suggested to the Council by Donald Laundry and Howard Zucker from the Columbia University College of Physicians and Surgeons, individual healthy stem cells would be extracted from 4- or 8-cell embryos that have failed to generate viable embryos for in vitro fertilization (IVF). In the second option, one or two cells would be removed from living 4- or 8-cell embryos and developed separately in culture. The third method, suggested by council member William Hurlbut from Stanford University, proposes the creation of embryo-like biological artifacts capable of developing into viable pluripotent stem cells but never into humans. Finally, somatic cells might be reprogrammed to return to a state of pluripotency. For each of the four proposed sources, the Council presents potential ethical concerns, comments on the scientific plausibility of the proposal, and considers whether or not the proposed technique for obtaining pluripotent stem cells is likely to be put into practice if it turns out to be feasible.

The first proposal suggests getting pluripotent stem cells from IVF embryos that have died in attempts to produce a child. Many IVF embryos stop dividing at the 4- to 8-cell stage. These are usually frozen, then thawed, to give the embryo a chance to resume cell division. If, however, the embryo fails to resume cell division within twenty-four hours of thawing, it is unlikely to begin again.

While most cells of such embryos are flawed, sometimes a few appear to be normal blastomeres. Laundry and Zucker acknowledge the difficulty of establishing a solid definition for organismic death of embryos, but propose that the lack of capacity for continued cell division, growth, and differentiation could be used as a working definition until more specific biochemical criteria are established. The paper makes note of their argument that removing these cells to culture pluripotent lines is ethically akin to taking organs donated with consent from the body of a person who has died in a car crash.

The doctors also note that the embryos must die independently of human decision, that is, they must be thawed to begin a second attempt at inducing division and then fail to divide, not start to divide and left to die. This rule was created to safeguard against the creation of many more embryos than are necessary for in vitro fertilization so that extras could be claimed for research. Due to the potential for abuse of IVF embryos, the Council finds the Laundry-Zucker proposal morally acceptable only if strict rules are implemented regarding the conditions under which cells can be removed from embryos for research purposes.

The Council goes on to question the scientific feasibility of the proposal, since at the time of the white paper’s publication it had not been determined whether pluripotent lines could be cultured from single blastomeres. The Council also notes that many scientists might frown upon the idea

of using stem cell lines derived from embryos flawed enough to stop dividing. It does, however, encourage acceptance of the possibility of using such cells, arguing that there is no way to know if blastomeres taken from so-called dead embryos are inferior to blastomeres taken from healthy embryos.

The paper next considers the proposal that one or a few cells be removed using biopsy at a time late enough that the loss of cells would not harm the embryo, but early enough that the cells taken would still be pluripotent. The fact that blastomeres are already extracted from in vitro embryos during pre-implantation genetic diagnosis (PGD) is considered, but the paper points to ethical opposition to PGD on the grounds that it doesn't explicitly benefit the embryo undergoing biopsy: its long-term effects on human embryos (if they exist) are unknown, and it is used to discriminate against embryos with genetic impairments. The paper opposes this proposal on ethical grounds because of the potentially large risk to embryos.

The Council also raises questions about the viability of the proposal. Citing a congressional history of opposition to federal funding for research related to in vitro fertilization, the Council notes that while Nicolai Strelchenko successfully derived human stem cells from embryos in the 8- to 24-cell phase, he destroyed the embryos in the process.

Subsequent research has demonstrated the scientific feasibility of both the first and second proposals. In November of 2006, Mateizel et al. published "Derivation of Human Embryonic Stem Cell Lines Obtained after IVF and after PGD for Monogenetic Disorders" in Human Reproduction, showing that it is possible to culture a pluripotent human stem cell line post-PGD. Around the same time, Irina Klimanskaya et al. published "Human Embryonic Stem Cell Lines Derived From Single Blastomeres" in the November 2006 issue of Nature, which demonstrated that healthy pluripotent lines could be cultured from individual blastomeres obtained from single-cell biopsy.

The third method for obtaining the desired cells is the suggestion by William Hurlbut that what he calls biological artifacts could be engineered by implanting modified somatic cell nuclei into oocytes. The proposed method is derived from somatic cell nuclear transfer (SCNT), which is used to create totipotent cells. Hurlbut suggests that the nucleus to be implanted could be genetically modified so that it could not form a totipotent cell. Though such a procedure might lead to the formation of pluripotent cells, he argues, a true embryo would never be formed. Therefore human life would not be destroyed in the generation of the pluripotent line.

The Council expresses significant opposition to this proposal, noting that if the nucleus to be implanted were modified it might form flawed pluripotent cells, and arguing that it would be so difficult to perfect Hurlbut's proposed method that scientists would be more likely to seek private funding for unrestricted work than to spend valuable time developing techniques for creating biological artifacts. Ultimately, the Council declares the method ethically unsuitable due, among other problems, to the potential for exploitation of vulnerable women to obtain oocytes for research, and the concern that such a method for obtaining pluripotent stem cells could lead to the creation of other ethically concerning pseudo-human artifacts.

The fourth and final proposal considered is that human somatic cells might be reprogrammed to dedifferentiate back into pluripotency. Noting the possibility of inducing such dedifferentiation using cytoplasmic factors from oocytes or existing pluripotent lines, the paper enthusiastically describes the extensive benefits of this proposed technique, pointing out that it may eventually be possible to culture stem cell lines from any human, which could be used for individualized therapies. The authors admit that developing this technique would be highly challenging from a scientific standpoint, but indicate that the first steps have already been taken in returning blood, liver, and muscle cells to a multipotent state. The council's only concern with the fourth proposal is that if dedifferentiation were taken too far the cell could become totipotent, effectively producing a clone and a new embryo.

Great leaps have been made in this area of study. Publications by Kazutoshi Takahashi and Shinya Yamanaka in 2006, Yu et al. in 2007, and Nokagawa, Koyanagi, along with others including Takahashi and Yamanaka in 2008, have shown that it is possible to induce adult somatic cells to return to a pluripotent state with retroviruses. In 2009 Hongyan Zhou et al. published " Generation of

Induced Pluripotent Stem Cells Using Recombinant Proteins," in which they explain how dedifferentiation of cells was achieved without the use of problematic retroviruses.

The white paper, "Alternative Sources of Human Pluripotent Stem Cells," provides a multifaceted examination of methods for obtaining pluripotent human stem cells that avoid destroying living human embryos. The majority of the paper is given to explaining the proposals and examining the potential ethical difficulties. The paper is significant because it gives voice to a variety of opinions–including several conflicting personal statements from Council members at the end of the paper. Even more important than the paper's internal debate, however, may be the way in which it seeks to influence further investigation. Since research involving destruction of embryos to create new pluripotent stem cell lines was ineligible for federal funding at the time this paper was published (a restriction removed by the Obama administration on 9 March 2009), this paper seeks to encourage research along specific pathways designed to produce pluripotent stem cells without the destruction of embryos. However, the fact that for several years foreign researchers have consistently published the most significant research on the fourth proposal–the proposal towards which the paper demonstrates the most favor–shows that the paper may not have convinced many American scientists to pursue the goal of government-sponsored human stem cell research.

Sources

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