Gene Therapy (2002) 9, 603–605

www.nature.com/gt

Editorial

Gene Therapy (2002) **9,** 603–605. DOI: 10.1038/sj/gt/3301769

The power to deliver: stem cells in gene therapy

Stem cell research has stimulated more media attention, public debate and government involvement than perhaps any other issue in the biological arena in living memory. This special issue of *Gene Therapy* comes at an opportune moment as scientists, patients and politicians join the debate on the potential of the twin technologies of stems and gene therapy for medicine in the twenty-first century.

The unexpected plasticity of differentiation displayed by stem cells from adult tissues has upset the dogma that tissue-specific cells are committed to a developmental fate. Until recently, a stem cell was by definition not differentiated, displayed a capacity for self-renewal throughout the lifetime of an organism, and had the potential to give rise to a large number of differentiated progeny. However, over recent times the potential of somatic stem cells for therapeutic applications has come to be viewed as almost infinite, limited only by the ingenuity of investigators in the manipulation of their genomes and culture conditions. Stem cell populations from some tissues may not be restricted to generating progeny identical to their origin, but instead have a plasticity that can be harnessed to generate cells of all germ layers. Allied to the enormous proliferative capacity of both embryonic and adult stem cells, this offers a wealth of opportunities for treatment and prevention of disease.

Still there remain significant lacunae in our understanding of the molecular mechanisms regulating adult stem cell development and differentiation. The sequencing of genomes from humans and experimental organisms followed by the advent of gene array and proteomic technologies has provided tremendous resources for stem cell research in the post-genomic era. New technological approaches make it possible to identify genes and proteins expressed in perhaps even individual cells. Characterisation of gene expression profiles in discrete stem cell subpopulations will facilitate our ability to identify the molecular mechanisms that regulate the self-renewal and differentiation of adult stem cells. Furthermore, expression profiling of adult stem cell populations from different tissues will provide important insights into the spectrum of genes which specify the identity and phenotype of a prototype stem cell. Such expression profiling will provide an essential tool in elucidating hierarchical relationships between stem cells and defining the molecular mechanisms that regulate stem cell plasticity. Armed with that knowledge, we might then be in a position to exploit the circuits controlling the biological processes for therapeutic purposes.

However, two recent studies suggest adult stem cells may not be quite as flexible as first thought.[1,2] Rather, the tissues they generate may arise from spontaneous fusion of the stem cells with other cells – which are then tetraploid – carrying unknown implications for the people who might receive them. Austin Smith, a stem cell researcher at the University of Edinburgh and a co-author of one of the studies, was quoted as saying that this 'suggests a need for caution with regard to the therapeutic use of adult tissue stem cells. If they only make other tissues by fusing with existing cells rather than producing new cells, their utility for tissue repair and regenerative medicine will be greatly restricted'. He added that 'if nothing else, our study indicates that calls for a halt to [embryonic stem] cell research are not scientifically justified'.

President Bush's stem cell policy rests largely on the premise that adult stem cells are versatile enough to produce all other tissues. Last summer, he issued regulations that would forbid federal funding of human embryonic stem cell research, except for cell colonies that already existed. The result is that only about 78 cell lines are available for federally funded research (Table 1).

As we go to press, the *Washington Post* has reported that the White House will officially nominate Elias Zerhouni, currently executive vice-dean of Johns Hopkins University School of Medicine, as the next director of the National Institutes of Health. President Bush is reported to have chosen Zerhouni to head the agency after assurances that he would support the administration's controversial limits on stem cell research and its support for a comprehensive ban on human cloning. In early March 2002, the Senate Health, Education, Labor and Pensions Committee held a hearing on the issue of therapeutic cloning. The star witness, actor Christopher Reeve, warned that the US would 'lose its pre-eminence in science and medicine' if therapeutic cloning was banned. But the most significant development was an indication that some committee members, including physician Bill Frist, may be getting ready to announce support for a ban on therapeutic cloning. Others want to limit applications

Table 1 List of laboratories holding existing stem cell lines

Name of laboratory	*Existing stem cell lines*
BresaGen, Inc., Athens, Georgia	4
CyThera, Inc., San Diego, California	9
Geron Corporation, Menlo Park,California	7
Karolinska Institute, Stockholm, Sweden	6
Maria Biotech Co. Ltd, Seoul, Korea	3
MizMedi Hospital - National University, Seoul Korea	1
National Centre for Biological Sciences/Tata Institute of Fundamental Research, Bangalore,India	3
Pochon CHA University, Seoul, Korea	2
ES Cell International, 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
Göteborg University, Göteborg, Sweden	19
Wisconsin Alumni Research Foundation, Madison, Wisconsin	5

Source: National Institutes of Health.

of technology still further, to include somatic cell nuclear transplantation. At a hearing of a US Senate Appropriations Subcommittee in March 2002, Gerald Fischbach, Dean of the Faculty of Medicine at Columbia University drew attention to the importation portion of the S.1899 bill introduced by Senator Brownback and others to amend the 2001 Prohibition of Human Cloning Act. This would enact criminal penalties against doctors and patients who seek to access treatments developed in other countries using nuclear transplantation. Similarly, an American who might travel to another nation to take advantage of a medical technology unavailable in the United States could be considered a criminal.

If such legislation were to be adopted, it would preclude the great potential for combining nuclear reprogramming with ES cell derivation to develop human pluripotent stem cells for cell-based gene and tissue therapies. ES cells derived from somatic cell transplantation could restore function to diseased or damaged tissues, or be genetically altered before transplantation to deliver gene therapy. Transplantation studies in mice have shown that ES cell-derived cardiomyocytes, neural precursors, haematopoietic precursors and insulin-secreting cells can survive and function in recipient animals. Several significant hurdles need to be overcome before this approach becomes feasible for clinical application. Not least is the identification of a suitable source of donor nuclei and the enhancement of nuclear reprogramming efficiency for so-called therapeutic cloning. In addition, robust pluripotent stem cell culture and *in vitro* differentiation systems are still a long way from being established. All these issues require intensive research to resolve for the benefit of patients worldwide. A recent report[3] in which completely immunodeficient mice were cured by transplant of ES cells genetically repaired by homologous recombination suggests that the principle of using somatic cell nuclear transfer to combine therapeutic cloning with gene therapy is sound, but much more needs to be done on translating this to clinical benefit.

The restrictions on stem cell research in the US will drive researchers and patients alike overseas in their quest to get the best from the technology. The UK Medical Research Council is proposing to set up the world's first human stem cell bank. The bank, which is likely to hold both adult and embryonic stem cell lines, will be overseen by an Advisory Committee chaired by Professor Alan McGregor (Kings College, London) with ethical, legal and regulatory issues coordinated by another committee chaired by Professor Genevra Richardson (Queen Mary College, London). Experiments on cloned and surplus human embryos were approved in February 2002 by the UK House of Lords (upper house of Government), which ruled that there were not enough ethical objections to outweigh the potential benefits to science and medicine. However, it stressed that the Human Fertilisation and Embryology Authority should not allow embryos to be cloned for stem cell research using cell nuclear replacement (CNR) unless there is a 'demonstrable and exceptional need'.

It has been suggested that embryonic stem cells may provoke less of an immune response than solid organ transplants – indeed, it has been speculated that expression of Fas ligand by ES-like cells might be a mechanism by which they can escape immune surveillance, allowing second-set allograft acceptance in experimental models.[4] However, this may not be true of the differentiated tissue derived from the embryonic stem cells.[5] Major histocompatibility complex antigen expression, and therefore immunogenicity, will depend upon the cell type into which the stem cells differentiate. Tissue derived *in vitro* from embryonic stem cells would probably lack endogenous antigen-presenting cell populations and could, theoretically, induce a weaker immune response, but the transplantation procedure itself would be likely to induce inflammation. The creation of a large pool of embryonic stem cell lines would increase the chances of matching MHC antigens. However, more than a million different lines would be required to create a comprehensive stem cell bank – well beyond the capacity of the panel of embryonic stem cell lines to which the US government has decided to restrict further support. An alternative is to make embryonic stem cells less immunogenic by eliminating or introducing surface antigens through genetic engineering, but this might lead to problems with genetic instability over time. Interestingly, although compared with somatic cells, embryonic stem cells develop a relatively small number of mutations – typically restricted to deletion of a chromosome that is replaced by a second copy of the remaining chromosome – they increase in frequency with time in culture.[6] Larry Goldstein, a cell biologist at the University of California (San Diego, CA, USA) said the finding that mutations accumulate in mouse stem cells in culture suggests that the 78 cell colonies approved for federal funding 'are not likely to be maximally beneficial for medical treatments'.

Derivation of ES cells from human sources by parthogenesis, if feasible, could circumvent some of the ethical concerns associated with human ES cell research. Recently, a stable macaque ES cell line was established from a parthenogenetically derived blastocyst (originating from a non-fertilized ovum) and could be induced to differentiate into a variety of cell types, including dopaminergic neurone-like cells, smooth mus-

cle cells, and spontaneously beating cardiomyocyte-like cells.[7] Up to 15 weeks after injection into the peritoneal cavity of severe combined immunodeficient (SCID) mice, derivatives of all three germ layers, including cartilage, neurones, hair follicles, and intestinal and respiratory epithelia were observed.

This issue of *Gene Therapy* highlights some of the most important progress that has been made in the understanding of stem cell biology and its exploitation for the treatment and prevention of human disease. We anticipate that gene therapists and those in the wider community of molecular medicine will find much to interest and excite them here. We look forward to a rich harvest from research in both basic and clinical applications.

NR Lemoine
Department of Cancer Medicine
Imperial College School of Medicine
Hammersmith Hospital
Du Cane Road, London, UK

References

1 Terada N *et al*. Bone marrow cells adopt phenotype of other cells by spontaneous cell fusion. *Nature* 2002; advanced online publication DOI: nature730.
2 Ying QL, Nichols J, Evans E, Smith AG. Changing potency by spontaneous fusion. *Nature* 2002; advanced online publication DOI: nature729.
3 Rideout III WM *et al*. Correction of a genetic defect by nuclear transplantation and combined cell and gene therapy. *Cell* 2002; published online, March 8.
4 Fandrich F *et al*. Preimplantation-stage stem cells induce long-term allogeneic graft acceptance without supplementary host conditioning. *Nat Med* 2002; **8**: 171–178.
5 Editorial: Accepting stem cells. *Nat Immunol* 2001; **2**: 1085–1086.
6 Cervantes RB *et al*. Embryonic stem cells and somatic cells differ in mutation frequency and type. *Proc Natl Acad Sci USA* 2002; **99**: 3586–3590.
7 Cibelli JB *et al*. Parthenogenetic stem cells in nonhuman primates. *Science* 2002; **295**: 819–820.