4 ‘Embryo-saving’ alternatives?

This Section deals with the search for ‘embryo-saving’ sources of embryonic stem cells that could be used in autologous cell therapy. ‘Embryo-saving’ means that no human embryos have to be created in order to obtain the stem cells, thus enabling skirting of the related moral and legal discussion.

Isolating stem cells from surplus embryos is also embryo-saving in this sense, but this approach does not produce stem cells with a tissue type that is an exact match for the patient. The question is therefore whether methods (forms of SCNT or alternatives to it) can be devised that produce embryonic stem cells for autologous cell therapy without creating human embryos in the process.

A recurring theme in almost all alternatives that come up for discussion here is the idea that a non-viable embryo is not an embryo. To that extent, this search is being conducted not only with technical and scientific means, but also with conceptual and philosophical means. The first answer (SCNT does not produce embryos) is itself based purely on this approach.

Clonotes

Though other mammals that developed from SCNT embryos were born after Dolly, they appear to be the exceptions. Most embryos die, but in most cases where an embryo does survive to birth the animal was found to have severe abnormalities related to faulty gene expression (Cibelli 2002a). It would seem that ‘clones that survive to birth merely represent the least abnormal animals’ (Jaenisch 2004). It has been demonstrated that these findings cannot be dismissed as ‘teething problems’ with reproductive cloning. Rather, they seem to indicate a possibly insurmountable biological barrier: the failure to reactivate essential embryonic genes that are marked as ‘switched off’ in the transplanted cell (Jaenisch 2003).

Some authors have recently concluded from this that it is incorrect to say that an SCNT embryo has the potential, just like an IVF embryo, to develop into a human being (Jaenisch 2004, McHugh 2004a, Trounson 2002). By implication, this would remove the right to protection conferred on human embryos and would open up the possibility of using these 'products of SCNT' (Jaenisch 2004) as sources of transplant material without further discussion of whether human embryos can be created for research or therapy. A shift in terminology has also been suggested: these entities should be called ‘clonotes’ rather than ‘zygotes’ or ‘embryos’ (McHugh 2004a). ‘We should use zygotes for babies, clonotes for cells’ (McHugh 2004b).

However, many IVF embryos are also lost *in vitro* or following implantation, which can often be attributed to chromosomal abnormalities. This means that the difference in viability between IVF and SCNT embryos is, at most, a question of degree. And how important is it to the debate as to the status of the embryo that all kinds of serious health problems are likely when an SCNT embryo does develop into a child (‘reproductive cloning’)? Do ‘potentially sick human beings’ have a different moral or legal status from ‘potential human beings’? It would seem difficult to uphold such an argument.

Cybrids

'Interspecies SCNT' involves transplanting a human somatic cell nucleus into a denucleated animal egg cell. There are two reasons for considering this procedure. First, it has been argued that this technique would probably not create human embryos, avoiding the issue of the moral and legal acceptability of the technique (Chang 2004, Solter 2003). Second, this would free SCNT from dependence on human donor egg cells. Of course, both advantages would only apply if the procedure delivered a usable source of human embryonic stem cells that were safe to use in therapeutic applications.

Little research has been carried out into this possibility so far, and the results have generally been disappointing. It is true that Chinese scientists recently reported that they had succeeded in obtaining human embryonic stem cells from embryos produced by transplanting human cell nuclei into denucleated rabbit cells (Chen 2003), but doubt has been cast on whether these claims are fully justified (Solter 2003). No evidence has yet been produced showing these cells to have all the properties needed for therapeutic use of embryonic stem cells.

The safety of the procedure relates to the possible risk of cross-infections. The chance of cross-infection is probably low, as the technique does not make use of the nucleus of the animal egg cell (where animal viruses might be found). More clarity is needed on this issue before therapeutic use of embryonic stem cells obtained via interspecies SCNT can be considered. Of course, this proviso does not apply to fundamental scientific research.

Assuming that interspecies SCNT can be a useable and safe source of stem cells, this proposal raises two issues (De Wert 2001). First, are the embryos created by this process human or not? We need to consider the hybrid, or more accurately 'cybrid', nature of such constructs: the nucleus of one cell-type combined with the cytoplasm of another (Solter 2003). Does the fact that part of the genetic material of a human-animal *cybrid* of this kind is of animal origin mean that we cannot regard it as a *human* embryo? One objection to this argument is that almost all the genetic material of such an embryo is derived from the nucleus of the human body cell used in the procedure (Lanza 1999, Health Council 2002, De Wert 2003). The animal input consists only of the cytoplasm with the mitochondrial DNA responsible for cellular energy balance (Health Council 2001).

Then we turn to the second question: if these human-animal *cybrids* have to be regarded as human, are they then also viable? Can these *cybrids* develop into human beings (with mitochondrial DNA of animal origin in all their cells)? If further (*in vitro* or animal) research shows that they are certainly not viable, then what is their ontological, moral and legal status? Some people consider that this raises the question as to whether they are actually *embryos*.

Embryo-like artefacts

Until it has been clearly established that *clonotes* and *cybrids* are definitely not viable, the moral implications of this for how they can be treated remain undecided. The approach described below is aimed at

resolving this uncertainty. A technical adjustment to the SCNT process would allow embryos to be created that do not have the potential to develop into human beings. This approach, 'creation of non-viable embryo-like artefacts', was put forward as an option for further investigation in a recent report of the US *President's Council on Bioethics* (The President's Council 2004)[1].

Dr. Hurlbut, an ethicist who is a member of this council, has suggested that this might be achieved by deliberately introducing a mutation into the nucleus of the cell body used in SCNT. He is thinking of a gene (cdx2) that is required immediately after the blastocyst stage (the stage at which stem cells can be harvested) to allow the placenta to form. If this gene is not correctly expressed, then the embryo will not be able to become implanted in the womb and so will not be able to develop into a human being. Dr. Hurlbut considers that a structure of this kind cannot really be regarded as an embryo and so can be created and then emptied without any objections being raised (Hurlbut 2004, Hurlbut 2005).

The scientific response to this suggestion has been divided. Dr. Melton, a biologist, raises the question of whether a genetic mutation of this kind has the same effect on human embryos as in mice, and wonders whether it would be possible to obtain useable stem cells from the artefacts produced. The energy and resources needed to investigate this aspect would be taken away from really significant research, and it is doubtful whether this approach would satisfy those who are opposed to research on human stem cells (Melton 2004). Others consider that it is worth the difficulty of giving it a try (Ready 2005).

Parthenotes

Parthenogenesis involves chemically stimulating an unfertilised egg cell to transform it into a diploid cell. This produces an embryo-like entity with two identical sets of maternal chromosomes. This process does not involve fertilisation or cloning. It is thus an alternative to SCNT rather than a variant of it. Of course, the 'uniparental embryos' or 'parthenotes' are female. They are also not viable, as no extra-embryonic tissue needed for implantation and further growth is created. However, parthenotes could perhaps be used as a source of embryonic stem cells.

There has in the meantime been success in obtaining parthenogenetic 'embryos' from research on mice and apes (Boediono 1999; Cibelli 2002b). These were shown to have the ability to develop into blastocysts, and researchers have succeeded in obtaining cell lines that do not appear to differ from cell lines from embryos produced by fertilisation. However, initial attempts to obtain embryonic stem cells from human parthenotes failed (Cibelli 2002b). Furthermore, this method would only produce cell material matching the tissue type of the woman from whom the egg cell was taken.

It has been suggested that parthenotes may not be human embryos as they have only 'partial generative potential' (Hurlbut 2004, Jochemsen 2004, PCB 2004). However, another argument is that parthenotes should be regarded as non-viable embryos with a moral status that remains to be determined (De Wert 2003).

1 A further 'White Paper' of the President's Council was published in May 2005, shortly after the Dutch original of this report went into print (The President's Council 2005). This provides a further discussion of 'alternative sources of human pluripotent stem cells', including the idea of creating embryo-like artefacts via SCNT and genetic modification referred to in this report.

Direct reprogramming

Another alternative to SCNT, in which there has been interest for some time, is a technique known as direct reprogramming. The aim of this method is to develop pluripotent cells (the equivalent of embryonic stem cells) from somatic cells, without having to create an embryo as an intermediate stage. This procedure is, in view of the advantages thought to be associated with it, regarded as *the* alternative for the future, (De Wert 2004, Trounson 2002). As with 'therapeutic cloning' via SCNT, this technique should produce matching tissue type transplant material for autologous cell therapy, thereby avoiding rejection. As no egg cells are needed for direct reprogramming and no embryos are produced, the moral objections raised against SCNT do not arise with this alternative.

But work on direct reprogramming is still in the very early stages and much more research is required. A first line of approach is trying to understand how (in SCNT) the denucleated egg cell or substances in the ooplasma (the cytoplasm of the egg cell) reprogramme the somatic cell nucleus. Once we know what mechanisms and substances are involved, we can try to imitate this in the laboratory via chemical induction. Much of this research can be conducted using animal material. However, species-specific differences mean that the question cannot be avoided of whether, and if so when, this research may need to be repeated on humans, creating *human* embryos via SCNT. In other words, this embryo-saving alternative can only be developed by means of research in which not only animal but also human embryos are created. It seems that this approach cannot, at least at this stage, help us get around the question of whether this is acceptable.

Another line of investigation might, indeed, allow this. This approach is looking into the possibility of direct reprogramming by transplanting a somatic cell nucleus (SCNT) into a human embryonic stem cell (Tada 2001; Do 2004). It appears that embryonic stem cells are capable of (at least partially) reprogramming somatic cells. This alternative would be embryo-saving (as embryonic stem cells rather than embryos are created) and also 'female-friendly'.