Opinion on the establishment of collections of human embryo cells and their use for therapeutic or scientific purposes

N°53 - March 11, 1997

Contents

Ever since its creation, questions have been put to the CCNE regarding ethical problems arising out of research using dead human embryonic and foetal samples. This was the subject of the Committee's first Opinion dated 22nd May 1984. The Opinion served subsequently as guidance for research programmes. A complement was added in the form of Opinion n° 23 dated 13th December 1990 concerning intracerebral transplants of mesencephalic tissue from human embryos to patients with Parkinson's disease for the purpose of therapeutic experimentation.

Law n° 94-654 of 29th July 1994 regarding donation and use of components and products of the human body, broaches the question of research on the embryo (article L. 152-8 of the Code of Public Health). The prohibition contained in the law, with the specific exceptions provided, protects all living human embryos. Research on embryos conceived *in vitro* and cultivated *ex vivo* are directly concerned by this text. Research on a dead embryo or foetus is not covered by the law.

Development of research requiring human embryonic sampling led to considering the possibility of creating human embryo cell collections and this prompted the CCNE to add a complement to Opinion n° 1.

Collections of human embryonic cells

I) - The possibility of growing and multiplying human cells *ex vivo* has expanded considerably since 1950 following research on viral diseases in humans since some viruses can only multiply on human cells. Human embryonic cells were chosen because they have remarkable proliferative potential *ex vivo* so that starting with a single embryo a large quantity of cells can be produced and frozen for storage.

These normal embryonic cell lines, after their characteristics had been very thoroughly verified, were made available to the entire scientific community by institutions such as W.H.O. and they are used as cellular bases for diagnoses, preparation of reagents (viral antigens) and primarily as media for industrial production of vaccines against viral diseases such as rubella and rabies.

Apart from these " normal" human embryo cell collections, there are also human cells from individuals affected by genetic diseases, or embryos from elective abortion following a antenatal diagnosis of such diseases. These cell collections have made it possible to conduct important research on the origins and mechanisms of hereditary disorders.

The cells which are cultivated are generally fibroblasts, or in some cases, lymphoblasts. These cells are not very differentiated but they are easy to cultivate and very long-lived. Differentiated cells however, generally have limited proliferative power and only short-term cultures are possible.

Introduction into human cells cultivated *ex vivo* of a gene of the oncogenic virus SV40 gives them an unlimited division potential. It has therefore been possible to create cell lines from certain organs, such as liver, kidney, cartilage.... Some of these lines are of embryonic origin and retain in a stable manner some of the characteristics of differentiation specific for the tissue they came from. These differentiated cell lines are particularly useful in cellular pharmacology research, and especially so in industry where they can sometimes replace animal models. Finally, some of these cultured cells are used, and in future may be increasingly used for therapeutic purposes. There are collections of differentiated cells created by research laboratories, or in some cases companies, who market them.

Collections of differentiated embryonic cells can also be created from frozen cells in view of later therapeutic use. This could be an option for grafting cells of the nervous system for transplants.

II) - Collections of a different type of human embryo cell could appear soon : **stem cells** , of which there are several kinds :

- **Tissue specific stem cells** . These are precursors of various cell populations constituting a differentiated tissue such as the hematopoietic system, the nervous system, muscles, etc. These cells can, in principle be used to try and reconstitute tissue which has been damaged by disease or development abnormality, but they do not in any way participate in the constitution of the germ cell lines, i.e. gametes.

- **Embryonic stem cells** , also called ES cells, are in principle " totipotent" . The cells were first established in mice starting with the inner cell mass of embryos in the blastocyst stage.

They are not in themselves either " embryos or eggs" since they are not independently capable of co-ordinated evolution to become a multicellular embryo and a normal foetus.

These cells however are totipotent in that they can participate in the formation of any tissue when they are injected into an authentic embryo in the morula or blastocyst stages, including the formation of germ cell lines. Mice created by these experiments are somatic and germinal chimeras, producing in particular gametes which are able to transmit the genome either of the embryo or of the ES cells.

- When they are cultivated *ex vivo* , these ES cell lines can either reproduce identically whilst retaining their totipotency with the help of experimental manipulations, or else differentiate into precursor cells of various somatic tissues and finally into differentiated cells. The type of differentiation can be controlled through culture conditions and various agents. As soon as ES cells begin to differentiate they lose their totipotency and can no longer contribute to the formation of germ cell lines.

More recently, embryonic stem cell lines have been established using the internal cellular mass of a ewe's and a rhesus monkey's blastocysts. Although the " totipotency" of these cells is not entirely confirmed, they could be heralding the forthcoming creation of human embryonic stem cells for which the field of potential applications is vast :

- furthering knowledge on cellular differentiation and on tumour formation

- creation of large quantities of differentiated cells which could be used as graft material to treat various diseases, blood disorders for instance, or of the immune system, the nervous system, or of muscles.

Such uses would require collections of human ES cell lines to be constituted, duly characterised (immunological phenotype, absence of chromosomal abnormalities or of contamination by infectious agents, or of transformation capacity).

* * *

Methods which can be put to biological use are continually generating new instruments, some of which carry truly therapeutic possibilities as well as difficult ethical problems.

This is particularly true of human embryonic stem cells which, although they may not exist as yet, could be established in the near future. Were the matter not previously discussed, difficult problems could face biologists, physicians, and public health authorities.

It is with this in mind that the CCNE wishes to formulate recommendations regarding the possible use of techniques and therapeutic instruments which have not yet been fully developed but which it is logical to believe might well be available very soon. The range of prospects and ethical issues brought about by human embryonic stem cells is such that the CCNE feels justified in preceding the event of which they are analysing the possible consequences.

Recommendations

A - Establishment of collections of cells using samples taken from a dead embryo or fetus after expulsion

The CCNE recommends the following :

1) - The terms of the ethical Opinion established by the CCNE on 22nd May, 1984, are maintained.

2) - The study protocol must be submitted by the research team with a presentation of cognitive or medical objectives, and by the obstetricians who will present the conditions in which sampling will be performed for consideration by the *Commission Nationale de Médecine et de Biologie de la Reproduction et du Diagnostic Prénatal.* (National Committee on Reproductive Medicine and Biology and on Antenatal Diagnosis).

3) - The patient's consent must be given after she has been informed both orally and in writing of the objectives of the research protocol which has been approved.

When general anaesthesia is necessary in order to perform elective abortion, it is preferable to obtain the patient's consent before operating so that information is not given at a time when a patient's powers of reflection might be impaired.

4) - In the case of elective abortion, information and consent cannot be considered before the legally defined reflection period has expired so that the knowledge that an embryo may be used for scientific purposes comes after the patient has taken a decision.

5) - Written material for consent must be kept for thirty years and filed securely to ensure total confidentiality.

B- Establishment and use of human totipotent stem cells from blastocysts

- Human stem cells of this kind, equivalent to ES cells in mice, do not exist as yet, but several laboratories outside France are working on their creation. Thus, the CCNE considers that its mission demands that it should as of now formulate recommendations on the conditions according to which they could, possibly, be established and used.

1) Article L. 152-8 of the Code of Public Health at present bans any embryo research ; because of this, the creation of ES cell lines from human blastocysts obtained by *in vitro* fertilisation and cultivated *ex vivo* is not possible.

However, taking into account important prospects in the field of therapeutic research, new articles might well be drafted when the law is up for revision at the end of 1999 which should make it possible to modify the ban.

2) With that in mind, only frozen embryos donated by couples who have given written consent, forsaken their parental project and decided to put an end to conservation, could be used for research.

3) However, any creation de novo of human embryos for any purpose other than a parental project, is still not permitted.

4) In the same spirit, collecting human embryos conceived *in vivo* by uterine lavage before transplant with the purpose of establishing cell lines, must be banned.

5) In conformity with recommendations set out in the CCNE's Opinion n° 8 of 15th December 1986 on " research and use of *in vitro* human embryos for medical purposes" , new arrangements must specify which kinds of research are admissible and which should be banned.

Use of human embryonic stem cells must be limited to :

- fundamental research activities

- or therapeutic research according to existing regulations.

6) In this connection, therapeutic applications which could modify the recipient's genome must be prohibited. Embryonic stem cells could be used to remedy somatic tissue deficiencies (for instance, grafting erythroid, or nerve, or muscle precursor cells), but this would need to be done in such a way that they could not in any event participate in the constitution of the germ line, i.e. male or female gametes and thereby transmitted to descendants.

7) Any use of ES cells with the aim of creating several human embryos with identical genomes must be banned.

In no circumstances should cells of human origin be used for commercial purposes regardless of whether they were obtained in France or from other countries in order to comply with principles which have been stated over and over again by the CCNE (Opinion n° 9 of 23rd February 1987) and incorporated in the law dated 29th July, 1994.

Comments concerning the status of "ES cells" and embryos conserved in vitro

Olivier de DINECHIN March 30, 1997

Even more so than the ontological status of embryos, the **ontological status of human ES cells** seems enigmatic. Their **origin** by sampling on a human embryo qualifies them as human cells. Their **totipotency** differentiates them from differentiated cells and puts them closer to the embryo. Could the fact that **it is necessary to intervene** , and more specifically to connect them to other embryonic material so that totipotency can be activated confer on them some similarity to gametes ?

As to their **ethical status** , it must take into consideration these two same facts : their origin and their totipotency. Their origin by sampling from a human embryo requires that the ethical status which is recognised for the embryo which they could produce be respected.

For these reasons, one might think that **their ethical status should be established at the same level** as that of the embryo. This means that it would be more demanding than the status of tissular stem cells or of other human cells.

It is for that matter what some of the recommendations adopted by the CCNE would tend to express : recommendation n° 2 requires authorisation to be given by the couple who produced the original embryos; recommendation n° 3 mentions " de novo" creation of human embryos (meaning implicitly inter alia the use of such cells); recommendation n° 6 limits use; and recommendation n° 5 refers to the CCNE's Opinion n° 8 on the subject of " research and use of in vitro human embryos for medical research.

oOo

The enigmatic presence of humanity in an embryo, albeit obtained and preserved in vitro, for which transfer is no longer a reasonable option, leads me to consider that there is an ethical difference between the two following attitudes towards such an embryo :

arrest conservation and therefore allow it to die naturally;

use it for research

This second attitude in fact reduces the embryo to the level of an object, or even to that of research material.

Limitations suggested by the CCNE regarding the use of in vitro embryos are essential in view of considerable demand and probable developments in research requirements. However, I do not feel they take account of this difference in sufficiently clear terms.

Report

on the constitution of human embryonic cells, tissues and organs collections and their use for scientific or therapeutic purposes

Ever since its creation, questions have been put to the CCNE regarding ethical problems arising out of research using dead human embryonic and foetal samples. This was the subject of the Committee's first Opinion dated 22nd May 1984. The proposals expressed in that Opinion have since served as guidance for research programmes. A complement was added in the form of Opinion n° 23 concerning intracerebral transplants of mesencephalic

tissue from human embryos to patients with Parkinson's disease for the purpose of therapeutic experimentation.

Law n° 94-654 of 29th July 1994 regarding donation and use of components and products of the human body, broaches the question of research on the embryo (article L. 152-8 of the Code of Public Health). The prohibition established by the law, with the specific exceptions provided, protects all living human embryos. Research on embryos *in vitro* are directly concerned by this text.

Research on a dead embryo or foetus is not covered by the law. An embryo or foetus expelled from the mother's body after implantation cannot survive before the 24th week of gestation. For such an embryo or foetus, a difference has to be made between death of the entire organism by cessation of the circulation of the blood supply brought about by expulsion from the mother's body, and cellular death since cells can remain viable for several hours. This cellular survival is one of the essential conditions for diagnostic, cognitive, or clinical research to be able to proceed.

Development of knowledge in these fields has led to considering the possibility of creating human embryo cell collections.

Ethical problems arising out of collections of embryo cells are not identical to those raised by collections of embryonic tissues and organs :

As regards cell collections , these remain viable and it is possible either to store the frozen cells or to create, using a single embryo, large quantities of cells cultivated *ex vivo* which are then kept frozen with a view to research and possibly medical applications.

In connection with tissue and organ collections, conservation techniques suppress viability and it would therefore be necessary to collect a great many embryo samples which would be available for cognitive research.

Embryo sampling possibilities

A) Embryos conceived in vitro before transplant in utero

Oocytes are fertilised *in vitro* and, by successive cellular divisions, embryos thus formed develop *ex vivo* to the morula stage (32 cells) made up of cells which are still sheathed in the oocyte's zona pelucida and then progress to the blastocyst stage when a cavity is formed. These cells (blastomeres) will then develop to the trophoblast (placental formation) or become the embryonic disc made up of cells which progressively will produce the various truly embryonic tissues. The embryonic disc will appear around the 14th day.

During this evolution, there will be a progression from a transient state of totipotent stem cells to various decisions for each cell moving gradually towards differentiation.

In medically assisted reproduction, embryos are inserted *in utero* when they are composed of two to eight cells (2nd to 3rd day).

If they continue to be cultured ex *vivo* beyond the blastocyst stage, transfer is no longer possible.

B) Dead embryo or fetus expelled after implantation

Expulsion can be either spontaneous or induced.

1) Spontaneous abortion

The high frequency of arrest of development after fertilisation is a well known fact. Only about one third of fertilised eggs develop to term. Most of these reproductive failures are eliminated during the first two weeks after fertilisation, before or after implantation and go unnoticed. Later, spontaneous abortions occur in about 25% of clinically recognised pregnancies, generally during the first trimester of gestation.

Research for purposes of diagnosis on spontaneous abortions have shown that the vast majority of them are the result of arrested embryonic development, consequence of a genetic anomaly, most frequently chromosomal. This fundamental fact must be kept in mind when research is contemplated based on cells whose source is spontaneous abortions.

Obstetrical data

More often than not, several weeks elapse between the time of arrested embryonic development and therefore of the death of the embryo, and the moment of spontaneous expulsion. The result is lysis of embryonic tissue. It is generally preferable for a woman's health to allow the process of spontaneous expulsion to take place in its own time and only induce evacuation if the mother's health were to be threatened by continued retention.

2) Elective abortion

We refer in this case to elective abortion practised within the terms of the law of 17th January 1975 (articles L. 162-14 of the code of public health).

- elective abortion before the 12th gestational week. The embryos are theoretically sound. (By international agreement, pregnancy is measured beginning on the first day of the last menses, i.e. about two weeks before fertilisation, so that the twelfth gestational week corresponds in fact to ten weeks of embryo development).

- therapeutic abortion motivated by the discovery of an abnormality which the law describes as " a condition of particular severity recognised as being incurable at the time of diagnosis" . Such anomalies are detected either through biological tests done on in utero samples of embryonic tissue, or by ultrasound examination. In such cases, the embryo or foetus is therefore affected by some disorder and this must be remembered if any cognitive research is envisaged using these cells. Clinical abortion carried out because the mother suffers from a serious condition is exceptional but in these cases, the embryo may be healthy.

Obstetrical data on elective abortion procedures

Elective abortion is performed either by the use of medication (Mifepristone or RU 486 and prostaglandins) and expulsion ensues without needing to use instruments (about a third of elective abortions) or by suction under general anaesthesia (see CCNE's Opinion n° 10 of 16th December 1987).

When embryonic cells are to be used for transplantation, the aspiration method is slightly modified. Instead of rapid aspiration which dilacerates tissue, manual gentle aspiration with a catheter under the guidance of ultrasound is used instead. This technique does not modify either the method or consequences of abortion for the mother, but does require a little more care on the part of the operator in order to preserve the embryo from undue damage.

Therapeutic abortion is generally practised at a stage of development when elective abortion techniques can no longer be used.

Some progress has been made in reducing the patient's sufferings and safeguard her obstetrical future. Medication is increasingly replacing methods formerly used. Pathological abnormalities which justify elective abortion must be verified by appropriate testing

beforehand so as to confirm or define the diagnosis with more precision. A cognitive research protocol using such cells must take that into account.

Criteria for determination of embryo death

Recommendation 1100 of the Council of Europe's Parliamentary Assembly discriminates between :

- a live pre-implantation embryo

-a dead pre-implantation embryo

-a post implantation embryo or a live foetus outside the uterus

-a dead embryo or foetus

Criteria used for defining death in a human cadaver for purposes of collecting organs (cerebral activity tested with an electroencephalogram) cannot be used for embryos.

The Parliamentary Assembly of the Council of Europe's document considers that death is determined by reference to the absence of vital functions such as spontaneous breathing and heart beats which in practice are not easy to observe.

An embryo or foetus which is expelled from the mother's body after implantation is not viable until it has reached about 24 gestational weeks. This is why the CCNE's Opinion n° 1 stated that " tissue sampling can only be performed on an embryo or foetus for which non-viability is a certainty, i.e. before the 22nd week of gestation (20th week after the probable date of conception)." The CCNE's Opinion went on to say that the absence of vital functions was determined by the arrest of blood circulation which at that stage of development is dependent on the mother's body.

Collections of human embryo cells

I . Since 1950 and following work by John Enders on the poliomyelitis virus, there has been a considerable extension of possibilities of cultivation and multiplying animal cells *ex vivo* . It was research on human viral diseases which led to an increase in demand for human cells cultivated *in vitro* since some viruses only proliferate on human cells. Human embryonic cells began to be in demand because of their copious proliferative capacity *in vitro* which makes it possible to produce a large number of cells.

Beginning in 1960, the Wistar Institute of Philadelphia (cell line WI38), and later in the London Medical Research (cell line MRC5) human embryo fibroblastic cell lines were developed, in both cases using an elective abortion embryo.

Cell freezing techniques made possible repeated use of the same stock of cells. After very stringent verification of their characteristics, these cell lines were made available to the scientific community by WHO and other similar institutions and were used for diagnosis, preparation of reagents (viral antigens) and above all as a substrate for production industrially of vaccines against viral diseases such as rubella and rabies.

Besides these collections of " normal" human embryo cells, there are also collections of human cells from genetically defective individuals or from embryos collected after elective abortion following antenatal diagnosis of these diseases. It has been possible to conduct important research on the origins and mechanisms of hereditary disorders using such collections. They are created and managed by laboratories (e.g. the biochemical laboratory in the Paul Brousse hospital in Lyons) or by institutions who make them available to research workers and ask for a financial contribution to cover operating costs of these "banks" (for instance, Corriel Institute in Camden, N.J.).

All of these collections are made up of fibroblastic cells , or sometimes lymphoblasts, which are not very differentiated but survive for a long time *in vitro.*

II. Differentiated cells have limited proliferative capacity and if no preventive measures are taken, their capacity for cultivation is short lived. This was the case in particular of kidneyl epithelial cells which were much in demand for virology research so that it was necessary to " collect" them on a regular basis and South Korean human embryo cells were commercialised as a result.

The introduction of an oncogenic virus gene (SV4O) in human cells cultivated *ex vivo* confers on them unlimited division potential. In this way, cell lines from certain organs such the liver, kidneys, or cartilage have been created. Some of these cell lines originated in embryos and retain in stable form some of the specific differentiation characteristics of the tissue they come from. These differentiated cell lines are particularly worthy of interest in cellular pharmacology research, in particular for industry where they can replace animal models. Finally, some of these cultivated cells are used, and in future may be increasingly used for therapeutic purposes. There are collections of differentiated cells created by research laboratories and in some cases companies who market them.

Collections of differentiated embryonic cells can also be created using frozen cells in view of later therapeutic use. This could be an option for grafting cells of the nervous system for transplants.

III . Collections of other varieties of human embryonic cells may appear in the near future, such as **stem cells** of which there are various kinds :

- Tissue specific stem cells . These are precursor cells of the differentiated cell populations which make up a differentiated tissue such as the hematopoietic system, the nervous system, muscles, etc. Such cells could, in principle, be used to repair tissue which has been damaged by disease or a development anomaly, but they do not in any way participate in the constitution of the germ cell line, that is the gametes.

- **Embryonic stem cells** , also called ES cells, are in principle " totipotent" . The cells were first established in mice starting with the inner cell mass of embryos in the blastocyst. They are not in themselves either " embryos or eggs" since they are not independently capable of coordinated evolution to become a multicellular embryo and a normal foetus. These cells however are totipotent in that they can participate in the formation of any tissue when they are injected into an authentic embryo in the morula or blastocyst stages, including the formation of germ cell lines. Mice created by these experiments are somatic and germinal chimeras, producing in particular gametes which are able to transmit the genome either of the embryo or of the ES cells.

- When they are cultivated *ex vivo* , these ES cell lines can either reproduce identically whilst retaining their totipotency with the help of experimental manipulations, or else differentiate into precursor cells of various somatic tissues and finally into differentiated cells. The type of differentiation can be controlled through culture conditions and various agents. As soon as ES cells begin to differentiate they lose their totipotency and can no longer contribute to the formation of germ cell lines.

More recently, embryonic stem cell lines have been established using the inner cellular mass of a ewe's and a rhesus monkey's blastocysts. Although the " totipotency" of these cells is not entirely confirmed, they could be heralding the forthcoming creation of human embryonic stem cells for which the field of potential applications is vast :

- furthering knowledge on cellular differentiation and on tumour formation

- creation of large quantities of differentiated cells which could be used as graft material to

treat various diseases, blood disorders for instance, or of the immune system, the nervous system, or of muscles.

Such uses would require collections of human ES cell lines to be constituted, duly characterised (immunological phenotype, absence of chromosomal abnormalities or of contamination by infectious agents, or of transformation capacity).

Should also be emphasised the possibility of applications which would obviously raise major ethical problems :

transfer of cell nuclei into enucleated oocytes and thereby opening the door to cloning (which has already been performed on domestic mammals)

injection of these cells, genetically modified or not, into human blastocysts in order to create human chimeras and therefore the possibility of transmitting the genome of these stem cells.

The possible value of therapeutic use of human embryonic stem cells and the eventuality of commercial use demand that further thought be devoted to the subject.

Collections of human embryonic tissue and organs

Research in the field of embryo genesis and development is important. Animal models, such as drosophila and mice, or fertilized eggs of the hen or quail, have made enormous contributions to progress in understanding fundamental data. Such data needs to be confirmed as regards human beings and for that purpose human embryonic material is required.

For embryonic tissue and organs to be stored, samples have to be fixated or frozen which destroys their viability. Only cognitive research can ensue.

Early this century, the Washington Carnegie Institute had made up a collection of embryos originating from spontaneous abortions. These samples were fixated and their examination gave rise to descriptions of the first phases in human development which were published in 1916. Later books on embryology were based on this publication. In 1920, again based on these same collections, a description of anomalies of embryonic development was published which highlighted the primary role of these anomalies as the origin of spontaneous abortion.

A few years ago, in the United States, the administration organised the creation of large tissue banks using samples collected after spontaneous abortions so as not to spark controversy about elective abortion. The programme was launched in spite of numerous warnings issued by the scientific community regarding its unrealistic nature because of the large number of genetic abnormalities causing early developmental arrest. It was dropped after only two years since less than 1% of samples could be used for research.

More recently, in September 1994, an NIH document prepared by the Human Embryo Research Panel, offered various possibilities for research on the human embryo. The House of Representatives rejected the conclusions of this report on 4th August 1995 and banned any human embryo research project with federal funding. There are no scientific or ethical recommendations as regards privately funded research.

Two situations exist in France

A. - Research for diagnostic purposes, done after elective abortion because a particularly severe abnormality in the embryo (or foetus) was discovered.

This is absolutely essential and the way in which it is performed will be the subject of implementation decrees to be applied to pluridisciplinary centres for antenatal diagnosis

(article 162-16 of law 94-654). Most pathologies causing abnormal embryonic development are uncommon and tests are carried out in very specialised laboratories which may set up small sample collections so as to further research on these subjects either in their own laboratory or in association with other researchers.

Collection of pathological human embryonic tissues or organs is not a problem in ethical terms and they are used in accordance with recommendations formulated by the CCNE's Opinion n° 1.

B - Research for cognitive purposes on normal embryonic tissue or organs.

Conditions in which such research is undertaken have, generally speaking, complied with recommendations in the CCNE's Opinion n° 1, and there are no other laws and regulations in force.

Research has been ongoing in recent years according to protocols submitted by research teams and by medical teams who are fully informed about the aims of the research programme.

In a few cases, research teams have joined forces to study embryonic tissues and organs which had not been used for a previous research project submitted by the scientific and obstetrical team. The scientific value of a new project using the samples should also be submitted to scientific authorities.

It has been suggested that, in the absence of any specific research project, collections of embryonic tissues and organs could be created by systematically harvesting normal but dead embryos from elective abortions so that tissues and organs could be made available to research units.

Normal tissue and organs can only be obtained from embryos following elective abortion within the framework of the law (articles 162-1 to 162-14 of the Code of Public Health).

Furthermore, as regards medical practices,

- embryo collection would require

Systematic co-operation on the part of hospital staff in charge of elective abortion facilities who would be asked to operate in technically more restrictive circumstances in the absence of any specific finality.

Attendance on a regular basis, in addition to medical staff, of technicians qualified to select, condition, and store specimens in such a way that they retain qualities needed for various test methods used for research.

The CCNE in its Opinion n° 1, had emphasised the exceptional nature of the use of embryos : " Exception must be the rule here so as to avoid use turning into pressure for widespread abortion and becoming just another routine procedure."

Ethical and legal data

I . For embryonic cell samples from a dead embryo or foetus, recommendations in the CCNE's Opinion n° 1 must be observed.

" Decision to undertake, and circumstances (i.e. timing and technique, etc...) of elective abortion must not in any way be influenced by possible or desired later use of the embryo or foetus. The termination technique must be selected on purely obstetrical criteria whilst taking care to preserve the mother's obstetrical future.

Total independence must be established and guaranteed, under the supervision of the Ethics Committee, between the medical team who perform elective abortion and those who could subsequently be using the embryo or foetus.

Only an embryo or foetus below the viability threshold, the decease of which has been previously established, shall be used for the above mentioned purposes."

Information and consent

In Opinion n° 1, the CCNE suggested that " the rule could be flexible and implementation could depend on the specific circumstances of abortion." " ... it would seem desirable to give the mother a veto instead of systematically demanding her consent." .

In the present report, the CCNE considers that :

Elective abortion is a grave action; it is traumatic both physically and psychologically for the patient. Information and consent procedures regarding possible use of embryonic cells must take that into consideration.

Information, given both orally and in writing must be clear about the aims of the research project which should have been approved by a Committee of Ethics.

In the case of elective abortion, information and consent cannot be accepted before the legal reflection time has expired so that the knowledge that an embryo may be used for scientific purposes comes after the patient has taken a decision.

When general anaesthesia is not required to perform elective abortion, consent can be requested after expulsion.

When general anaesthesia is necessary to perform elective abortion, it is preferable to obtain the patient's consent before operating because if information were given later, her powers of reflection might be impaired or disturbed.

In any event, total confidentiality must be ensured and written consent documentation should be kept apart from medical files.

Oral information should be accompanied by a written document specifying :

1. . that use of specimens is only a possibility. It is not possible to predict the condition of the cells after expulsion and whether they can in fact be used.

2.the aims of the research programme and that it has been approved by an Ethics Committee.

3.rules of confidentiality.

II . For totipotent stem cells which are the blastomeres of the embryo *in vitro* , as it is presently formulated, the law forbids cell sampling .

Article L. 152-8 of the Code of Public Health states that :

" *In vitro* conception of human embryos for the purposes of study, research, or experimentation is prohibited.

" Any experimentation on embryos is prohibited.

" Exceptionally, a man and woman forming the couple concerned, may accept that their embryos be studied.

" Their decision is expressed in writing.

" Such studies must have medical objectives and must not be detrimental to the embryo.

" They can only be undertaken after regulatory approval has been granted by the Commission referred to in article L. 184-3 hereunder according to conditions defined by decree in the *Conseil d'Etat* .

" The Commission publishes an annual list of establishments where such studies are ongoing as well as their purpose."

Cell sampling in order to create collections would be detrimental to the embryo and is unacceptable as an application of this text.

Although using embryonic cells and tissues is not considered by laws n° 94-653 relating to respect for the human body, n° 94-654 relating to donation and use of elements and products of the human body, and n° 94-452 (Chapter IV, Products of gene and cell therapy), some articles included by these texts in the Civil Code, the Code of Public Health, and the Code of Intellectual Property, could apply to collections of embryonic cells created for research or therapeutic purposes (cell grafts for instance).

These articles concern :

1) The principle of non-patrinomy

- art. 16-1 of the Civil Code : " The human body, its elements and products cannot be the subject of estate law"

- art. 16-5 of the Civil Code : " Any convention whose effect is to confer property value to the human body, its elements or products is null and void"

- art. 16-6 of the Civil Code : " No payment can be made for allowing experimentation on one's person, or the taking of elements of one's body or the collection of products of that body"

2) Non patentability

- art. L. 611-17 of the Code of Intellectual Property. " The human body, its elements and its products as well as knowledge of the total or partial structure of a human gene, cannot as such be the object of a patent.

3) Anonymity

- art. 16-8 of the Civil Code and L. 665-14 of the Code of Public Health state " No information whereby can be identified either the person who has donated an element or product of his/her body or the beneficiary of the donation can be divulged. A waiver to this principle of anonymity can only be granted in case of medical necessity."

4) Consent

- art. L. 665-11 of the Code of Public Health : " Samples of elements of the human body and collection of its products cannot take place without prior consent of the donor. Such consent can be rescinded at any time.

5) Safety and hygiene regulations

- art. L. 665-15 of the Code of Public Health

6) Regulations concerning conservation, conditioning, use, distribution and disposal of tissues and cells

- art. L. 672-10, L. 672-11 and L. 672-12 of the Code of Public Health.

Article L. 672-11 is abrogated by law 96-452 of 28th May 1966 which redefines the status of products of gene and cell therapy.

In all cases article L. 145-16-1 of this law regulates " samples for the purpose of composing a collection of human biological specimens" . The expression " collection" means assembling, with a view to genetic research, biological samples taken from a group of persons identified and selected according to clinical and biological characteristics in one or several members of the group, and any by-product of such samples."

Are collections of embryo cells covered by this law if genetic research is involved?

Applying all these texts to collections of embryo cells is not a simple matter and ethical issues raised by the possibility of establishing human embryonic stem cell lines must also be taken into account.

Any research on the establishment of such cell lines is impossible in the light of article L. 158-8 of the Code of Public Health since it would be necessary to obtain cells from a blastocyst which would possibly have been kept under cultivation *ex vivo* beyond the time when it could be transferred.

Research teams who wish to work in this field which is of particular importance as regards scientific knowledge and possible therapy, can obtain these cell lines from abroad, in some cases on a commercial basis.

Are human embryo cells covered by decree 96-327 of 16th April 1996 and its texts of implementation of the same date published officially (in the *Journal Officiel*) on 18th April 1996 with entry into force on the 19th April 1996 ?

This new text on importing and exporting organs, tissues, and cells of the human body, lays down the following principles :

1) Donor consent and anonymity.

The establishment of nameless documentation which can however facilitate determination of a product donor is allowable for reasons of safety and hygiene.

2) Non payment of donations.

3) Importing or exporting tissue or cells for scientific purposes is subject to authorisation delivered by the Minister of Health.

Since this decree did not mention human embryo cells, an important ethical issue is the production of embryos *in vitro* with the sole aim of creating cell lines for collections be they for commercial purposes or otherwise.

We are approaching paradoxical situations as a result of legislation :

- there is a ban on research which can be detrimental to an embryo *in vitro* and therefore on research which could destroy it, but it can be destroyed after it has been kept for more than five years.

- experimentation or therapeutic research on totipotent cells from embryos in vitro are

banned, but it is possible to import cells from collections established without any observance of specific ethical laws applicable in France to embryonic cells.