Research Licence Interim Inspection Report

Project Title	Evaluation of Cardiomyocytes from Embryonic Stem Cells as a Means to Characterise Receptor / Channel Expression in Human Tissue
Centre Name	NURTURE
Centre Number	0076
Research licence Number	R0141
Centre Address	Floor B East Block Queens Medical Centre Nottingham NG7 2UH
Treatment centres donating to this research project	NURTURE Centre 0076
Inspection date	5 December 2007
Licence Committee Date	TBC
Inspector(s)	Debra Bloor
Person Responsible	Bruce Campbell
Nominal Licensee	Ian Johnson
Licence expiry date	31 May 2009

About the inspection:

The purpose of the inspection is to ensure that centres are carrying out research in compliance with the HF&E Act 1990, Code of Practice, licence conditions and directions.

The report is used to summarise the findings of the inspection highlighting areas of firm compliance and good practice, as well as areas where further improvement is required to improve patient services and meet regulatory standards. It is primarily written for the Licence Committee who make the decision about the centre's licence renewal application. The report is also available to patients and the public following the Licence Committee meeting.

This report covers the period between April 2006 and December 2007.

Brief Description of the project

Lay summary

Embryonic stem cells can be formed from blastocyst-stage embryos and have the ability to form any cell of the body e.g. nerve cells, heart muscle, and liver. Recent breakthroughs in stem cell technology has increased the possibility of producing cells that could be used to treat many types of degenerative diseases such as Parkinson's, muscular dystrophy and liver failure. Additionally human stem cells could be potentially used for

- Research on the mechanisms controlling the transformation of embryonic stem cells into different cell types, e.g. heart muscle cells (known as cardiomyocytes);
- Screening new drugs to determine their therapeutic value and potential toxicity.

Human stem cells can form heart muscle cells that can be observed to contract (beat) in the culture dish. This is controlled by receptors for compounds such as adrenalin. Any changes in the pathways for these receptors are associated with severe cardiovascular disease. These receptors also control the activity of smooth muscle cells including those in the airways of the lung where contraction (spasm) leads to asthma. The current project focuses on receptor systems in heart and smooth muscle cells formed from stem cells to assess whether these cells behave normally.

Research activities	Research on human embryos	✓
	Storage of licensed material	
	Creation of embryos for research	
	Derivation of human embryonic stem cells	✓
	Cell nuclear replacement	

Changes/ improvements since last inspection

In April 2007 the stem cell derivation team moved to purpose-built laboratories in the Wolfson Centre for Stem Cells, Tissue Engineering and Modeling. The move doubled the capacity for human embryonic stem cell culture, with the new facility housing 8 class two cabinets, including two purpose-built for stem cell derivation with in-built dissecting microscopes.

Although the stem cell derivation laboratories have been relocated, it is intended to continue to culture embryos to blastocyst stage in the laboratories of NURTURE prior to their transfer to the new laboratories.

The new laboratories in the Wolfson Centre were visited in October 2007. The Wolfson Centre is based in a separate building from NURTURE and in consideration of the requirements of the 1990 Human fertilisation and Embryology Act[1] the centre was advised that stem cell derivation should not take place in the new laboratories until the licence has been reviewed by a Licence Committee.

Additional licence conditions and recommendations and actions taken by centre since last inspection

The licence was issued without any additional conditions.

Recommendations of the 2006 interim report

Recommendation	Action taken
The donating centre's method for completing the HFEA treatment registry forms was not consistent. In most cases these forms only stated that embryos had been used in research if the embryos had been grown to blastocysts and passed into the research laboratory. It was recommended that if embryos are placed in extended culture with the intention of using them to derive embryonic stem cells then these embryos should be classed as having been used in research and reported as such to the HFEA.	Reporting procedures have been revised to comply with the recommendation.

[1] At paragraph 4 (2) (d) of schedule 2 the 1990 Human Fertilisation and Embryology Act it states that a licence cannot apply to premises of the person who holds the licence in different places.

Recommendations of Licence Committee considering 2006 interim report

Recommendation	Action taken
The director of embryology at NURTURE is involved both in the research project and in the clinical care of patients. The Committee agreed that the centre should be reminded to ensure that the person who speaks to patients about donating material for treatment is not involved with their clinical treatment. They also agreed that the centre should explore ways of making the distinction between research and treatment as clear as possible. In particular, the importance of maintaining this distinction should be clearly emphasised in the centre's protocols.	The director of embryology confirmed that on the rare occasions that she is involved in clinical procedures relating to a patient who has consented to the donation of material to research that she would not participate in decisions related to the selection of embryos for the clinical treatment of the patient.
The Committee noted the patient information for the project does not contain any mention of the fact that a copy of patient consent forms will be sent to the national stem cell bank with any hES line deposited. This needs to be rectified. The Committee also identified the following additional problems with the patient information: a) The Committee agreed that the words "UK Government" should be removed from the second paragraph under the heading "What rights do I have over the stem cells made from my spare embryos". b) Under the heading "What if I change my mind", the Committee agreed that more clarification should be provided about when embryonic material is deemed to have been used in the research. c) The Committee also agreed that a customised consent form should be made available to patients who had material in storage which was approaching the end of its storage period.	On the basis of comments made on the suitability of patient information made in the last inspection report and by the Licence Committee that considered the report, it was reported that the unit reverted to the use of an alternative, previously approved form of patient information. It was reported that a copy of a patient information leaflet used by the human embryonic stem cell consortium is being reviewed with the intention of it being used in the future.

Summary for Licence Committee

The current research project has been in progress since 2004. The centre has suitably qualified and experienced staff and the premises are secure and appropriately equipped. All patients donating material to the project consented to participation in the research.

A number of issues for consideration were identified in the course of the inspection and these related to the following areas of practice:

- Compliance with the terms of the HFEA licence;
- Ethical committee approval for the research;
- The documentation and reporting of donation to the research project.

As a result of the relocation of laboratories where it is proposed to carry out stem cell derivation the project has not been fully operational since April 2007. As the new laboratories are in a separate building from the licensed premises, the licence committee is asked to consider whether a second licence is needed for the stem cell laboratories.

The inspector would support the continuation of the licence conditional on the resolution of the licensing of the stem cell derivation laboratories and NHS research ethics committee approval for the research should donations continue to be accepted from patients receiving NHS funded treatment.

Proposed licence variations

The focus of the research has changed since the inception of the project in 2004 (see page 14). The Licence Committee is asked to consider whether the lay summary of the project should be varied to reflect the current focus of the research more accurately or whether the change in focus is sufficient to warrant relicensing of the project including further peer review of the proposed research.

Report of Inspection findings

1. Organisation

Desired Outcome: The centre is well-organised and managed and complies with the requirements of the HFE Act.

Summary of findings from inspection
Evidence of:

- Leadership and management
- Organisation of the centre
- Resource management
- Staffing
- Funding

Staff

Person Responsible	Bruce Campbell
Principal investigator	Lorraine Young
Scientists	Christian Denning, Cecelia Sjöblom, Sarah Chamberlain, Maria Barbadillo-Munoz
Support staff (receptionists, record managers, quality and risk managers etc)	Staff from centre 0076

Highlighted areas of firm compliance
The person responsible is suitable qualified and experienced and has been the licence holder since the inception of the project in 2004. In the time since the licence was renewed, the scientist who derived the centre's two cell lines joined NURTURE to train as an embryologist. She has continued to assist the senior scientific officer working on the derivation laboratories in assessing embryos in the first few days after they are plated onto feeder cells. The senior scientific officer from the research team has also been designated as laboratory manager. Laboratory procedures for stem cell derivation are documented and risks associated with the procedures have been assessed. The laboratory manager also has responsibility for staff training. Although no new staff have been recruited to work on the licensed research project in the time since the last inspection, the laboratory manager was able to provide evidence that other staff in the team undergo induction training and further training in specific procedures and that the provision of this training is documented. It was reported that this training would be provided to any new members of the team working on the licensed research.

Issues for consideration
The stem cell laboratory has no formal funding for the stem cell derivation work undertaken and continues to obtain consents and attempt derivations at a relatively slow rate. Prior to the inspection of the Wolfson Centre laboratories in October 2007 an embryo was transferred to the new laboratories for attempted derivation of stem cells. The laboratories of the Wolfson Centre are in a separate building to that of the licensed premises of NURTURE: paragraph 4 (2) (d) of schedule 2 the 1990 Human Fertilisation and Embryology Act states that a licence cannot apply to premises of the person who holds the licence in different places and in consideration of this, transfer of the embryos to the Wolfson Centre constituted a breach of paragraph 3 (1A) (a) of the 1990 Human Fertilisation and Embryology Act (HF&E Act). Section 3 (1A) (a) states that no person shall keep or use an embryo, except in pursuance of a licence. It is also stated at paragraph 4 (1) (a) of schedule 2 of the Act that a licence under this Schedule can only authorise activities to be carried on premises specified in the licence. No licensed research has taken place on the premises of the Wolfson Centre subsequent to the inspection in October and the incident was reported to the HFEA. The PR should review how the requirements of the HFEA licence are disseminated to all members of the research team. It is also recommended that a policy for the reporting of adverse incidents is developed in line with the requirements of standard S 9.4.1 of the 7th Code of Practice (COP) and that when this is complete, adverse incident reporting requirements are communicated to all members of the team. A six monthly progress report was submitted to the HFEA in November 2007. This is the first report that has been submitted following the renewal of the licence in May 2006. The licence has not been fully operational over the last year and it is noted that the PR considered that as a result of the temporary suspension of the research, no progress report was required. Although newly recruited members of the research team are provided with health and safety training in the course of their routine induction, staff of the research team appeared unfamiliar with the requirements to participate in annual mandatory health and safety training. It is recommended that the PR seek the advice of the local health and safety representative to establish what annual training is required and that he ensures that all members of the research team participate in the relevant training as required. Participation in the training should be recorded. The team expressed reservations about the security of consent forms held by the secretary of the steering committee of the National Stem Cell Bank (NSCB). It is recommended that the PR seeks guidance and reassurances from the NSCB on the security of consents held by the NSCB and that a robust system is put in place to ensure that the team can comply with the requirements of standard licence condition A 19.6 (e). In the course of the inspection it was reported that when the initial research application was made, approval for the project was obtained from the NURTURE local ethics committee. At that time, the patients donating material to the project were largely self funded. However, NURTURE now provides NHS funded treatment to a significant proportion of its patients.

Advice provided on the National Patients Safety Agency website states that ethical advice from the appropriate NHS research ethics committee is required for any research proposal involving patients and users of the NHS[2]. In consideration of this the PR reported that a decision has been made to apply for NHS research ethics committee approval for the research. If appropriate the PR should consider suspending donation by patients whose treatment is funded by the NHS until appropriate approval is obtained.
Executive recommendations for Licence Committee
Since the initial stages of the research take place on the premises of NURTURE and paragraph 4 (2) (d) of schedule 2 the 1990 Human Fertilisation and Embryology Act states that a licence cannot apply to premises of the person who holds the licence in different places the Licence Committee is asked to consider whether a second licence is required for the laboratories of the Wolfson Centre.
Areas not covered in this inspection
Research governance

[2] http://www.nres.npsa.nhs.uk/applicants/apply/research-in-the-nhs/

2. Premises and equipment

Desired Outcome: The premises and equipment are safe, secure and suitable for their purpose.

Summary of findings from inspection:

- Suitability of premises
- Storage facilities
- Safety of equipment
- Servicing and maintenance of equipment

Highlighted areas of firm compliance
In the initial stages of the research embryos are cultured on the premises of NURTURE (centre 0076) in secure laboratories accessible by licensed personnel only. In the course of a routine interim inspection of the treatment and storage licence of centre 0076 in November 2007 evidence of maintenance of a sample of key laboratory equipment was observed and it was reported that the laboratory had been subject to a local health and safety inspection. The cryostore facilities were inspected and were considered suitable. Frozen embryos are not stored under the auspices of the research licence but remain in storage under the auspices of the treatment and storage licence of centre 0076 until they are thawed and used in the research project. The centre was considered to have a robust bring forward system for the identification of material reaching the end of its consented storage period. The cryostore in the laboratories of the Wolfson Centre is fitted with a low oxygen level alarm (no embryos are stored under the auspices of the research licence). The cryostore is fitted with a ventilation system that is linked to the alarm. Evidence of the routine maintenance of class II cabinets in these facilities was seen in the course of the inspection and it was reported that other equipment is monitored and cleaned regularly. It was reported that the laboratories in the Wolfson Centre have been subject to a local health and safety inspection although no formal report of the inspection was available. In the report of the 2006 interim inspection it was noted that HFEA treatment forms were annotated to indicate that embryos had been used in research only if the embryos subject to extended culture reached the blastocyst stage of development. It was recommended that if the embryos were placed in extended culture with the intention of using them to derive embryonic stem cells then these embryos should be classed as having been used in research and reported as such to the HFEA. A review of information held in the HFEA register and comparison of this information against records held by centre 0076 showed that this recommendation has been adopted.
Issues for consideration
Access to the laboratories in Wolfson Centre is controlled but the laboratories are shared facilities and non licensed personnel do have access to the area. It was reported that should research on these premises be licensed, viable embryos will not be left unattended before being plated onto feeder cells and that after plating, embryos are no longer considered viable. It was recommended that before any derivations are carried out in the Wolfson Centre, protocols are revised to reflect that viable embryos should not be left unattended in shared laboratory areas.

Inactivation of the low oxygen level alarm in the Wolfson Centre requires access to the cryostore. It is recommended that the advice of the local health and safety representative is sought to establish how the alarm can be deactivated without exposing staff to a potentially oxygen depleted atmosphere and that warning signs are displayed to advise staff not to enter the laboratories when an alarm is sounding.
Executive recommendations for Licence Committee
The Licence Committee is asked to consider the proposed practice of culturing embryos in the process of becoming attached to feeder cell layers in laboratories that could potentially be accessible to non licensed personnel.
Areas not covered in this inspection
None

3. Donation of material

Desired outcome: Ensure donors are recruited in a proper way and their consent is respected.

Summary of findings from inspection: *(Delete areas not being reported on)*

- Prevention of coercion of prospective donors
- Ensuring patient consent is not breached
- Donor and patient records

Highlighted areas of firm compliance
The records of all the patients who donated embryos to the research in the time since the last inspection were reviewed. Specific consents to the participation in stem cell research were present in all the records. It remains the case that the director of embryology of NURTURE plays an active role in the research project. Following up concerns raised in the previous inspection report that there could be a perceived conflict in these roles, the director of embryology confirmed that on the rare occasions that she is involved in clinical procedures relating to a patient who has consented to the donation of material to research that she would not participate in decisions related to the selection of embryos for the clinical treatment of the patient.
Issues for consideration
There were some discrepancies between the number of embryos reported as donated to research in the 6 monthly progress report, the number recorded as donated in HFEA register records and in the centres own records of donated material. This is potentially a breach of standard S.7.3.2 of the COP[3]. In the course of the inspection the records of all patients donating embryos to research in the time since the last inspection were reviewed and on the basis of what was recorded in the records all the discrepancies were resolved. However, the PR should review the procedures for recording the donation of embryos to research and, in liaison with the PR for centre 0076, for the accurate completion of HFEA treatment forms. Members of the embryology team at centre 0076 (also included on the research licence of project R0141) agreed to ensure that the required corrections are made to HFEA treatment forms. While it is acknowledged that the director of embryology does clearly distinguish between her clinical and research roles, it remains a recommendation that laboratory SOPs are revised to reflect the practices designed to ensure that there is no conflict in these roles.
Executive recommendations for Licence Committee
None

[3] S.7.3.2 of the COP states that the procedures for traceability of gametes and embryos shall also ensure that registers are kept of received, processed, stored and distributed or discarded gametes or embryos, enabling identification of (c) distributed gametes or embryos and hospitals or institutions to which gametes or embryos have been distributed (whether intended for application in the human body, or research purposes).

Areas not covered in this inspection
Recruitment of donors Ensuring prospective donors have access to further guidance Ensuring prospective donors have time to consider donation properly These areas of practice were reviewed in the course of the previous inspection when they were considered largely appropriate.

4. Patient information and consents

Desired outcome: Ensure that patients are informed in order to give informed consent

Summary of findings from inspection: *(Delete areas not being reported on)*

- Patient information
- Patient information for projects deriving embryonic stem cells
- Consent forms for projects deriving embryonic stem cells

Highlighted areas of firm compliance
With one exception (detailed below) all of the recommendations of the previous licence committee relating to patient information and consents were adopted.
Issues for consideration
On the basis of comments made on the suitability of patient information made in the last inspection report and by the Licence Committee that considered the report, it was reported that the unit used an alternative, previously approved form of patient information. It was reported that a copy of a patient information leaflet used by the human embryonic stem cell consortium is being reviewed with the intention of it being used in the future.
Executive recommendations for Licence Committee
None
Areas not covered in this inspection
None

5. Scientific practice

Desired outcome: Procedures are robust to ensure material is used appropriately

Summary of findings from inspection:

- Minimisation of material loss and wastage
- Progress towards achieving set aims and objectives

Use of material
In the time period between 6 February 2006 and 23 November 2007 21 fresh and 7 frozen embryos have been donated to the project. All of the fresh embryos and one of the frozen embryos were used in the research. Six of the frozen embryos donated to the project remain in storage. The donated embryos were cultured and 5 formed blastocysts which were transferred from NURTURE to the (old) stem cell laboratories for derivation attempts and 1 was frozen for a future derivation attempt. Of the fresh embryos used for derivation attempts, no embryos formed stem cell lines. One frozen embryo did not survive the freeze thaw process. The project has used fewer embryos than anticipated but it was reported that patient recruitment has been slower than expected. In addition, the stem cell facility has been out of action for some time for development and refurbishment and pending consideration of the status of the stem cell derivation laboratories by the HFEA.
Project objectives
1. Establish methods for the derivation, propagation and differentiation of human embryonic stem cells into cardiomyocytes; 2. Evaluate alternate methods for the propagation, directed differentiation and isolation of cardiomyocytes and smooth muscle cells from human ES cells: 4. Determine the effect of source, passage number and method of derivation, propagation and differentiation on the characteristics of muscle cells derived from human ES cells. It was reported that the research undertaken to date has concentrated on Objectives 1, 2 and 4 in terms of the derivation of hESC lines (Objective 1) and differentiation into cardiomyocytes (Objective 2) and the effect of using medium containing physiological levels of methyl group donors on epigenetic instability (Objective 4). Objective 3 of the original application aimed to “Characterise β-adrenergic and muscarinic receptors in cardiomyocytes and smooth muscle cells derived from hESCs in relation to other in vitro model systems.” As the project has progressed studies reporting these evaluations have appeared in the literature and interest in pursuing this objective has waned. It is therefore expected that the main experimental focus of the project will be in relation to Objectives 1 and 4 of the original application although Dr Chris Denning’s work on cardiomyocytes is ongoing. Three papers have been published detailing the results of the research.[4]

[4] Burridge P.W. *et al,* Stem Cells 2007 Apr;25(4): 929-38; Allegrucci C. *et al,* Hum Mol Genet. 2007 May 15;16(10): 1253-68; Kim K.P. *et al,* Genome Res. 2007 Nov 7; [Epub ahead of print] PMID: 17989250

Lay summary of research undertaken
Surplus embryos donated for research were cultured in NURTURE and transferred to the stem cell laboratory for derivation of human embryonic stem cell lines. Of these donated embryos, two embryos formed stem cell lines, named Nott1 and Nott2 and these lines have been characterised and submitted to the UK Stem Cell Bank. These lines have formed the basis of three scientific publications so far and work is ongoing to examine the effects of the use of more physiological media during derivation of the lines on the normality of the cell lines obtained
Issues for consideration
When the project was originally licensed in 2004, it was customary for research projects to specify the purpose for which derived stem cells were to be derived and this is reflected in the detail of the original objectives. When the licence was granted, the focus of downstream analysis was on receptor systems in heart and smooth muscle cells derived from stem cells. In the course of the work it has been noted that hESCs appear to be genetically unstable and that instabilities arise during early passage of the derived stem cell lines. The current focus of the work has been varied to test the hypothesis that components of the culture medium used during the initial stages of derivation may impact on the genetic stability and the differentiation potential of resulting stem cell lines. The PR should consider revising the lay summary to more accurately reflect the focus of the work and the Licence Committee may wish to consider whether the focus of the research is sufficiently changed to warrant variation of the licence.
Executive recommendations for Licence Committee
None
Areas not covered in this inspection
Standard operating procedures Quality assurance systems

Report compiled by:

Name……Debra Bloor…….

Designation……Inspector……..

Date…18 December 2007……

Appendix A: Centre Staff interviewed

The PR and four other members of the research team met with the HFEA inspector

Appendix B: Licence history for previous 3 years

Licence R0141: Evaluation of Cardiomyocytes from Embryonic Stem Cells as a Means to Characterise Receptor / Channel in Human Tissue

Licence	Active from	Expiry date
R0141/3/a	01/06/2006	31/05/2009
R0141/2/a	01/06/2005	31/05/2006
R0141/1/b	01/03/2005	31/05/2005
R0141/1/a	01/03/2004	28/02/2005

All of the licences listed above were issued without additional conditions or recommendations.

The centre held an additional five research licences between 1995 and 2003.

Appendix C: Response of Person Responsible to the report

Centre Number……0076……

Name of PR…….Bruce Campbell ……

Date of Inspection…….5th December 2007……

Date of Response……7th February 2008……

Please state any actions you have taken or are planning to take following the inspection with time scales

The actions taken to comply with the report are dependent on the licence committee's decision with regard to the need for an additional licence for the STEM laboratory.
Prof. Young, the PI for this project, has indicated that the requirement to obtain an additional license, including in particular an NHS ethical approval, would represent a significant investment in time and resources that could not be justified in terms of the potential benefits gained and the time frames involved. Further, the current lack of a specific funding source for this project means that we would be unable to fully complete the current ethics form. We have therefore come to a decision to discontinue this project if the licence committee decides that we will require two licences (as appears likely). Given that the project has produced two stem cell lines that have been lodged in the stem cell bank and contributed to at least three scientific publications, we feel that this work will have made a significant contribution and the time will have come to draw it to a close.
Alternatively, if it is decided that we do not require an additional licence then it is likely that we will continue with this project and with the guidance of our HFEA inspector, I will instigate a series of actions to satisfy any regulatory recommendations made by the licence committee.

I have read the inspection report and agree to meet the requirements of the report.

Name……Bruce Campbell ……

Date……7th February 2008 ……

2. Correction of factual inaccuracies

Please let us know of any factual corrections that you believe need to be made (NB we will make any alterations to the report where there are factual inaccuracies. Any other comments about the inspection report will be appended to the report).

These matters have been discussed with Dr Bloor by Telecon on 7th February 2007 and no further corrections are required.

We also welcome comments about the inspection on the inspection feedback form, a copy of which should have been handed out at the inspection. If you require a copy of the feedback form, please let us know.

Please return this section of the report electronically to Debra.Bloor@HFEA.gov.uk or in hard copy to:

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