***SUPPLEMENTARY INFORMATION***

***Experimental Medicine Cell Bank Service***

**Rationale: Ensuring Bio-integrity**

The division of Experimental Medicine has an excellent research profile that spans from basic science to translational medicine, “from bench to bedside” and is critical to develop enhanced diagnostics and treatments for human disease. The research within the department has an esteemed reputation and a track record of excellence, which should be maintained at all costs. One of the ways to ensure Experimental Medicine remains a fore-runner of world class research is to develop laboratory services that are novel and meet the current and predicted working practices put forth by collaborating institutions, government regulators, funding agencies and scientific peer-reviewed journals.

 The creation of a departmental cell bank should take into consideration the information presented above to ensure the highest standards are applied during the design stages to benefit the future. In my opinion, the ideal design of a departmental cell bank would not be limited to physical storage of numerous cryopreserved cell lines but as a complete service provided to the members of Experimental Medicine. This service would be designed and managed by the Laboratory Management Office where all processes would be performed according to standard operating protocols by the Laboratory Management Team. In this manner, this comprehensive service will ensure the highest quality of product released back into circulation and retain the biological integrity of the samples from registration to release. This service will further support the researcher’s desires to expand the experimental portfolio of the division and the University of Oxford.

**Design Plan**

**Operators**

As stated previously, this is a service that will be operated only by members of the Laboratory Management Team to ensure bio-integrity. The tissue culture operations will be performed primarily by the Laboratory Assistant who will be responsible for thawing registered vials, growth and expansion of cell lines and preparing cell lines for freezing. All processes will be covered by standard operating protocols and a log book for observations.

The Assistant Laboratory Manager will be responsible for entering the registration data, overseeing the processes of tissue culture work with associated documentation and assisting if necessary, and taking control of frozen cell lines and transferring to long term storage.

The Laboratory Manager will oversee the general process of the cell bank, periodically checking that SOPs are followed and performing annual audits of the cell bank. The Laboratory Manager will also liaise with Group Leaders regarding permissions, levels of stock and the option of authentication testing by an outside source.

**Facilities**

Experimental Medicine Division currently has two cryogenic facilities, room 5603 and 7607. The cell lines operating under the departmental cell bank would primary be held in the newly purchased LABS 20K cryostorage tank supplied with a dedicated liquid nitrogen supply by a 240 litre tank in room 5603. 10% of the cell stocks will be stored in the 10K cryostorage tank which also has a dedicated liquid nitrogen supply by a 180 litre tank, also located in 5603.

Tissue culture activities for the cell bank, ideally should be performed in a small tissue culture suite with restricted access to individuals only undertaking cell bank activities to ensure strict, good laboratory practice and guarantee a sterile, controlled environment.

The ideal, proposed area for these activities to be performed is in laboratory 7402C (previously Radiation suite) where there is limited access, a functional class II microbiological safety cabinet, cell counting station with an inverted microscope, centrifuge and room for an CO2 incubator to be installed.

This space has gradually been made suitable for this purpose but still requires removal of old equipment and integration of CO2 incubators to create the ideal tissue culture suite.

 **Inventory Management**

The cell bank inventory will be managed using a specialist inventory control system software package (currently under investigation).

This system will allow the operators to generate a unique identifier upon registration, record all sample details, produce working stocks from the master stock, record the inventory location, any associated test results and accompanied documentation (ex. authenticity certificates).

All samples stored within the LABS 20K cryostorage tank must be bar-coded. This will enable any storage box to be checked quickly and accurately. The use of bar-coding for samples will be made available for all users to request storage in this tank, separately from the cell bank samples.

Access to inventory locations or sample data will only be by the Laboratory Management team but will be made available with restrictions, any other users of the software package. This system will be fully audited annually by the Laboratory Manager.

**Process**

All procedures will be followed according to standard operating procedures written by the Laboratory Manager using similar guidelines to that of the required procedures for the Human Tissue Authority. Certain, critical procedures may require witnessed signatures by two operators to ensure quality control of the samples. These SOPs will be available to view by all Group Leaders who have banked cells. A brief flowchart of the entire process can be viewed in appendix A.

***Registration***

The first step is to register a cell line to the cell bank. Upon completion of the registration form, the frozen vial of the line will be physically transferred to the Lab Management -80°C Freezer store to begin processing (within a week, if longer, it will be stored in liquid nitrogen). Details required for registration are;

 Name

 Group Leader

 Laboratory

 Cell Line Name

 Species

 **Cell Line Origin (Collaborating Laboratory, ATCC, ECACC, etc…)**

 Passage Number

 **Release Permissions (No release, Open release, Restricted release)**

 Information pertaining to the **origin of the cell line**, whether it is from collaboration, uniquely created or purchased directly from a company, allows the sample to be coded as verified or un-verified. Verified samples are those that have been purchased directly from a company which provide authentication documents to certify the origins and genetic profile for the sample. The associated authentication documents would be added to the inventory control software and the integrity and “proven” origins retained with the sample. Unverified samples are those that have not been purchased directly from a company or received from a collaborator where genetic verification has not been confirmed with the registered sample.

 Registration of cell lines by a Principle Investigator, or an individual on behalf of the Investigator may wish to set **release permissions** on the samples submitted. For example, if a new cell line has been created by a member of the PI’s group and wishes to hand the sample over to the cell bank, but under the instruction that no other group has permission to use it, the line can be registered as **No Release**. Alternatively, the PI may wish to register the sample as **Open Release** if it is a well known cell line that has been verified and allow all within the department to use it. **Restricted Release** means the sample is available to another group upon approval obtained from the owner via notification through the Laboratory Manager.

***Preparation of Master Stocks***

Once the sample cell line has been registered and entered into the database, the sample will be thawed and resuscitated to grow and expand 50-100x106 cells to ensure cryopreservation of 25 master stock vials. Growth will begin in a T25 or T75 flask, upon reaching 90% confluency, the cells will be then transferred to T175 flasks to expand out the line further. By using a stepwise approach for expansion, the cells will receive adequate space and nutrition to grow and proliferate, however passages will be kept to the absolute minimum to preserve the genetic integrity and physical characteristics of the cryopreserved line.

 Once these master stocks have been made, one vial will be kept aside to grow and expand out a working stock. All master stocks will be frozen before a working stock is resuscitated to ensure controlled conditions.

 ***Preparation of Working Stocks***

A single Master stock vial will be thawed and grown out to create 25 vials of cells (50-100x106). The procedure is similar to above and then the vials will be frozen down at a controlled rate to ensure healthy cells upon resuscitation.

 ***Mycoplasma Testing***

Master stocks and Working stocks will be Mycoplasma tested using the Lonza MycoAlert kit during the growth and expansion phase of production.

 ***Release***

Release will occur by way of a notification to the Group Leader or nominated representative to say that the line has successfully been expanded and banked. The group then receives a single vial of working stock line to locally use and maintain.

***Optional: Authentication***

For registered, un-verified cell lines, there is an available option in which the Laboratory Management Office will organise shipment of a vial to a company to perform STR profiling to confirm the samples identity and genetics. This optional part of the service will be performed at cost to the Group (approximately £190) and all documentation will be attached to all cell lines generated after this verification.

**Intellectual Property**

The cell bank is an operated service which is under the remittance of Laboratory Management but the ownership of the individual cell lines will remain the property of the Group it was registered by. Any request for further information, permission and material transfer agreement requests will be forwarded directly to the Group Leader. Since these cell lines are assets to individual groups, they may fall under the category of Micro-organisms that may need to be recorded on the Annual Biological Returns; this is under the responsibility of the Group Leader. If requested, Laboratory Management can provide a report of cell lines and vial numbers to ensure accurate recording.

**Risk Management**

**Contamination**

Good laboratory practice will be followed and SOPs strictly adhered to at all time when performing any tissue culture procedures. The Laboratory Management Team has received ample cell culture training to ensure the highest quality work and results. Also, by tissue culture work only performed by one or two individuals who will record all work performed, any contamination can be traced back its source and investigated. In addition, master and working stocks will be tested for mycoplasma before being cryopreserved.

Contamination by other cell lines (cross-contamination) will be prevented implementation of only working with one cell line at a time. If additional cell lines need to be worked on within the same day, the more robust, quicker growing lines will be dealt with after working with the slower growing ones.

**Equipment Failure**

The samples will be kept in the new LABS 20K tank that has been ordered specifically for the creation of a cell bank. The supply of liquid nitrogen comes from a dedicated 240 litre liquid nitrogen supply tank which is not disconnected unless refilling and is only performed by the Laboratory Management team. The cryogenic facilities are checked daily by the Laboratory Management Team but in addition, there is an audible alarm for low liquid nitrogen supply. As a contingency, 10% of each sample type will be stored in a second departmental cryogenic tank, as a backup. The second tank is supplied by a separate, dedicated liquid nitrogen supply which is also checked daily.

**Loss of samples/Inaccurate inventory**

An inventory control software be used for the cell bank, which will include the use of barcodes and specialist laboratory labels which can withstand extreme temperatures to avoid deterioration of the label on the tubes. The inventory control system will be updated from registration, expansion, creation of master and working stocks, storage and release by members of the Laboratory Management team according to SOPs to ensure complete sample tracking. The software will also enable the team to view and audit all changes to the inventory to ensure a full audit trail can be recovered. To ensure quality assurance, a full audit will take place annually. This will include taking a sample vial from the tank to trace back to lot numbers of reagents used and registration documents located as well as choosing a registration and following the audit trail to locate the working stocks in the cryostorage tank as well as the contingency vials located in the alternate cryostorage tank. All vials stored within the cryostorage tank will be required to have a bar-code to ensure all tubes can be identified.