NCI-FREDERICK
INSTITUTIONAL BIOSAFETY COMMITTEE
MINUTES

OCTOBER 18, 2011

CALL TO ORDER / ANNOUNCEMENTS

The NCI-Frederick Institutional Biosafety Committee was convened at 12:10 p.m. in Building 549 Executive Board Room with the following members in attendance:

Voting (Quorum = 9)

- ☒ Michael Baseler
- ☒ Theresa Bell
- ☒ Rev. David Betzner
- ☒ Stephen Creekmore
- ☒ Bruce Crise
- ☐ Eric Freed (regrets)
- ☒ Melinda Hollingshead
- ☐ Stephen Hughes (regrets)
- ☒ Sarah Hooper
- ☒ Scott Jendrek (arrive 12:15pm)
- ☒ Serguei Kozlov
- ☒ Dan McVicar (Chair)
- ☒ Randall Morin
- ☐ Shalini Oberdoerffer
- ☒ Raja Sriperumbudur
- ☐ Lucien Winegar

Non-Voting

- ☒ Walter Hubert
- ☒ Kim DiGiandomenico

APPROVAL OF MINUTES FROM SEPTEMBER 20, 2011 MEETING

The minutes from the September 20th meeting were tabled for vote until the November meeting. Several members had problems opening the file from SharePoint and could not review them.

ACCIDENT REVIEWS

Accident reviews from August through the November IBC date were presented by Theresa Bell to the committee. In August, an investigator had a needlestick after injecting an animal. SOPs were being followed; however, it was determined that the needle was not disposed of immediately after injection, which led to the accident. No biological materials were involved in the incident. The individual was retrained by senior staff.

In September, an investigator was harvesting cells from the colon tissue of a mouse with a scalpel and subsequently cut her finger when scraping the blade on the top of an Eppendorf tube. The SOP was being followed; however after re-evaluation of the procedure, the investigator was advised to try a plastic (cell) scraper for the harvest, rather than a scalpel, to avoid future accidents.

Additionally, an animal care taker was bitten by an overly-aggressive mouse during a cage change. Appropriate SOPs were being followed at the time. The mouse was a 'clean' nude mouse from a breeder colony and no biologicals were involved.

Lastly, a lab worker was cut with an unprotected scalpel that was left inside a drawer while she was cleaning the laboratory. The cut was severe enough to warrant stitches. The PI retrained all laboratory staff on proper storage and disposal of sharps in the laboratory.

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REVIEW OF PROTOCOLS

NEW REGISTRATIONS

Lea Cunningham 11-50: *Evaluation of Benzodiazepine Ro-5-3335 Pharmacokinetics and Leukemic Efficacy in a Murine Leukemia Model* - Exempt per NIH Guidelines from full committee review-use of transgenic murine cell line ***Notification to committee***

Teizo Yoshimura 11-51: *Generation of mice deficient of beta defensin 4 and 14* BREEDING ONLY ***Notification to committee***

Robert Fisher 11-52: *Purification of Elongation Factor 2 from primary tumor cells for a study conducted by Dr. Ira Pastan (NCI)* To purify protein from human leukemia cells (ALL), cells will be lysed by high salt concentration, then by hypotonic shock. Water soluble extracted proteins will be loaded on small column containing ion-exchange resin and then eluted by 0.1 M salt. Then proteins will be precipitated by 5 fold volume of acetone. The precipitate will be dissolved in SDS-containing buffer and fractionated by gel electrophoresis. Gel stained and protein band digested by protease for mass spectrometric analysis. During the pre-review, there were some questions regarding what steps were done within a BSC. All steps up to and including the acetone step will be conducted following BSL2 practices and conducted within a BSC. Pending clarification in which room the BSC is located, Serguei Kozlov motioned to approve the registry. Bruce Crise seconded the motion. For: 12 Against: 0 Abstain: 0 *(Follow-up note: BSC is located in room 245)*

RENEWALS

Kathrin Muegge 11-48 (07-48): *Reprogramming of murine cells* The goal of this renewal is to express four genes (KLF4, Oct4, Sox2, myc) in murine cell lines and primary cell cultures. The transfected cells will be selected and examined for epigenetic changes. Because they need to express multiple genes simultaneously and because primary cultures are difficult to transfect, they need to use either a retroviral packaging system or a lentivirus packaging system. The four stem cell genes will reprogram the fibroblasts and will lead to induced pluripotent cells (IPS). Those IPS cells will be tested *in vitro* for induction of pluripotency markers (using e.g. real time PCR analysis, western or immunostaining). The group will examine the efficiency of reprogramming comparing wild type cells with Lsh-/- cells. Lsh is an epigenetic regulator that controls DNA methylation in mammals. During the pre-review, it was not clear if animal work would be conducted and there were questions on decontamination of wastes; however, all concerns were addressed sufficiently prior to full committee review. Theresa Bell motioned to approve the registry as written. Mike Baseler seconded the motion. For: 12 Against: 0 Abstain: 0

Snorri Thorgeirsson 11-53 (08-87): *Generation of mice hemizygous for MxCre transgene and homozygous for the TGF-beta II receptor (TBIIR) and c-met floxed allele* BREEDING ONLY ***Notification to committee***

Ulrich Baxa 11-54 (08-54): *Analysis of virus particles, cells and tissue samples by electron microscopy/ shared service* The Electron Microscopy Laboratory (EML) provides service to over 100 investigators in NCI-Frederick, NCI-Bethesda, and other institutes at the NIH and other government agencies. The service is performed as part of ATP's shared service, technology development, and collaborative effort. Possible samples are cell cultures, animal and human tissues, microorganisms, and virus preparations. All samples that are received by the EML are chemically inactivated by 4% formaldehyde and/or 2% glutaraldehyde at the researcher's laboratory for at least 2 hours, but usually 24 hours. Other less common workflows are negative staining of samples and cryo-electron microscopy. During the pre-review there were concerns regarding who actually does the fixation step and the validation of the inactivated tissue samples received. The EML has standardized fixation protocols that they provide to PI's who wish to use the services of the EML. EML also provides the reagents for the protocols to provide a level of consistency with the fixation step. To date no fresh, frozen samples of brain tissue have been received, only samples already fixed and embedded in paraffin. Theresa Bell motioned to approve the registration at BSL1 with the stipulation that the lab never receives fresh, frozen brain tissue. Bruce Crise seconded the motion. For: 12 Against: 0 Abstain: 0

Dimiter Dimitrov 11-55 (08-21): *HIV Entry and Neutralization* The major goal of this renewed project is to elucidate mechanisms of HIV entry and neutralization and to test a number of antibodies and other reagents for the inhibitory activity. The lab will also test fusion activity of cells infected with HIV. There were some questions during the pre-review regarding the SOP and where certain procedures would take place that were addressed. During full committee review, there were concerns about the TZM assay and inactivation of HIV. Dan McVicar moved to conditionally approve the registry pending clarification of the lysis buffer; more specifically if there is any detergent that at cell-mediated cytotoxic response (CMC) would not inactivate HIV. Brue Crise seconded the motion. For: 12 Against: 0 Abstain: 0 *(Follow-up note: Per PI response and review from virologists on IBC, lysis buffer should completely lyse cells as well as virus)*

Jonathan Keller 11-56 (08-01): *Transduction of human hematopoietic cells and cell lines with replication incompetent lentiviral vectors* The objective of the proposed work is to continue to investigate the function and regulation of genes which control blood cell development. Specifically, the genes proposed for delivery by lentivectors in this registration are tumor suppressors, or short hairpin RNA (shRNA) coding sequences designed to target potentially oncogenic proteins. The only exception to this is Id1, which appears to be mitogenic, the human coding sequence which was cloned into a commercially purchased expression vector. There was some discussion regarding the vector system. Even though the cells are pseudotyped with VSV-g, the vector system is HIV-based and cannot reassemble due to a large section of *env* that is deleted. Bruce Crise added that the genome is not mobilizable. Due to the nature of the vector and its application in the registry (to target potentially oncogenic proteins), the committee also highly recommended for new staff to conduct a 'dry run' of the experiment as part of their training to ensure proficiency of techniques prior to being permitted to handle the actual agents. Steve Creekmore motioned to approve the renewal with the aforementioned recommendation. Bruce Crise seconded the motion. For: 12 Against: 0 Abstain: 0

OUTSTANDING ITEMS

Christopher Westlake 11-49: *Functional Characterization of membrane trafficking genes in primary cilia and tumorigenesis* Originally deferred at the September meeting. After meeting with the PI and assisting with the document revisions, Dan McVicar requested expedited review of the revised IBC registration. Lead reviewers concurred and the revised documents were circulated via email for committee review. For: 10 Against: 0 Abstain: 0

Ji Ming Wang 11-43 (08-53): *The role of mouse G protein receptor FPR2 in infection, inflammation and cancer* Deferred and Listeria project suspended in September. Concerns with the paperwork have been addressed; however, the mock observation could not be conducted prior to October IBC meeting due to PI being out of the country. Due to another potential violation that occurred prior to the IBC meeting, the IBC requested for that employees access to the animal facility to be revoked, effective immediately. *(Follow-up note: Access was reinstated as of October 21, 2011)*

Trinchieri/Noer – IBC 10-60 Flow cytometry core lab: Waiting on response from PI/Project Supervisor

R. Gorelick 11-03-A1 – Lead reviewer requested full committee review – Hepatitis C Virus work – At September meeting, committee deferred request for additional information and concurrent submission with Julian Bess for his part in the project. PI met with Theresa Bell and Kim DiGiandomenico on October 17, 2011. Additional updates/information will be provided for review at the November IBC meeting.

AMENDMENTS

Seventeen amendments were processed between the September and October meetings. Six remained outstanding as of the meeting date, while eleven were approved.

OTHER BUSINESS

Kim DiGiandomenico provided the committee with an update on the revisions being made to the IBC website regarding biological fact sheets and tech bulletins. Ms. DiGiandomenico also requested feedback on the revised membership appointment policy which will be voted on at the November meeting.

ADJOURNMENT

The meeting was adjourned at 1:50 pm.

Next meetings: ***November 15, 2011*** ***December 20, 2011***