



**NCI-FREDERICK
INSTITUTIONAL BIOSAFETY COMMITTEE**

Minutes
June 19, 2007
NCI-Frederick

The NCI-Frederick Institutional Biosafety Committee was convened at 12:05 p.m. in the Building 549 Executive Boardroom with the following members in attendance:

Ms. Theresa Bell, Secretary	Dr. Henry Hearn
Dr. Stephen Hughes	Mr. Lucien Winegar
Ms. Alberta Peugeot	Dr. Mike Baseler
Dr. Bruce Crise	Dr. Melinda Hollingshead
Ms. Dianna Boissey	Dr. Stephen Creekmore
Dr. Dan McVicar	Dr. Jeanne Herring
Dr. David Garfinkel	

Members not in attendance: Dr. Randall Morin

Others in attendance: Ms. Cara Leitch, Dr. Scott Keimig, Dr. Robert Thomas, Dr. Charmaine Richman.

INTRODUCTION

Ms. Bell distributed the April 2007 minutes for review. An email vote will be taken early next week to document changes and final approval.

Ms. Bell asked the committee for comments on the Delinquency Statement to address non-compliant Principal Investigators who have delays in obtaining IBC approval. The committee reviewed the draft document distributed for the second time, with a modification to include the addition of transgenic, knock-out or genetically modified animals and a notation citing the approved revised IBC Charter signed on May 18, 2007. The committee had no further comments or requests for changes and the Delinquency Statement was approved.

Ms. Bell asked the committee for comments regarding the Supplemental Strain Forms used to consolidate key information particular to research involving animals when

multiple strains are used by an investigator. The committee reviewed the document, and with no comments or requests for further modification, the committee approved the form as an attachment for registration purposes.

PROTOCOL REVIEWS

RENEWALS

07-32 (Dr. Hu)

- Clarify the research scope and exactly what will be done and how.
- B5g: Define what exactly the standard cloning procedures include.
- The Standard Operating Procedure requires clarification (sandals, shorts, dresses, and the use of autoclave bags to store PPE, etc...).
- The use of BSL-2 or BSL-2* environments seems appropriately planned, dependent on the safety profile of the virus involved. Clarify areas defined as BSL-2 versus BSL-2* and which virus work will occur in these areas. EHS/IBC will conduct an inspection and smoke test of the area to confirm that the proposed work area is acceptable for the work scope.
- A4a: If any of the material is potentially infectious, then there should not be any work with sharps, to include cutting of gels or any other sharps related activities.
- Experiments are planned for up to 4L scales at times. For what specific viruses and at what titers does the group anticipate for the 4L scale (>10mL) experiments? What are the typical ranges of viral titers for their 0.1-10.0 mL lots?
- D9: Are FACS assays or sorting activities performed? What are the safety and containment procedures in this case, if applicable?
- Are new viruses that are supposed to be replication incompetent or impaired routinely tested to verify this property?
- Inactivation procedures in the SOP describe the use of 70% ethanol. Why is this being used in lieu of bleach or other more effective decontaminants?
- Is data available on the inactivation of the viruses being used or spill clean-up procedures?
- How often are lab coats replaced or cleaned? (The SOP states that lab coats are stored in autoclave bags on a hook.)
- Because of the use of MMLV and murine-based cells, provide a summary statement to address knowledge of the potential for mobilizable elements and how this risk will be addressed.
- Acknowledge the infectious potential of the DNA and how this risk will be mitigated.
- The committee raised the issue of appropriate footwear in the laboratory. Ms. Bell will research the issue and report back to the committee in July.

Ms. Bell made a motion to defer approval, Dr. Baseler seconded and all were in favor.

07-37 (Dr. Pathak)

- A1: Where are the PBMCs coming from (e.g. NIH)?
- A3: Clarify areas defined as BSL-2 versus BSL-2* (are these labs labeled correctly in the registration?) Which virus work will occur in these areas? EHS/IBC will need to do

an inspection and smoke test of the area to confirm the proposed work area is acceptable for the work scope.

- A3: How will the HIV-1 patient samples be handled safely? Address how MMLV and murine based cells will be handled safely.
- A6: This should be checked yes, since question A6a. was answered.
- B5b: Current specific viral genes in plasmids are described. How will future plasmid constructs be discussed with safety? How are the identities of the plasmids verified?
- C12a: Organisms are concentrated using filtration. Are liquids containing viruses pumped under pressure?
- C13a: Sterilization of outside of tubing is performed with ethanol. Why is bleach or other more effective disinfectant not used?
- D9: Are FACS assays or sorting activities performed? What are the safety and containment procedures in this case, if applicable?
- Are new viruses that are supposed to be replication incompetent or impaired, routinely tested to verify this property?
- The Standard Operating Procedure requires more detail (sandals and shorts are not recommended in the BSL2* lab.
- Clarify the use of autoclave bags to store PPE, and how often are lab coats replaced or cleaned?
- Inactivation procedures in the SOP describe the use of 70% ethanol. Why is this being used in lieu of bleach or other more effective decontaminants?
- Is inactivation of material verified during spill clean-up procedures?

Dr. Creekmore made a motion to defer approval pending resolution of the above items, Dr. Hollingshead seconded and all were in favor.

07-33 (Dr. Hornung)

- B8: appears to be contradictory. Hypodermic needles and syringes are to be avoided, but procedures for handling and disposal of needles are described. Previously sharps have been excluded (as indicated in A4). This should be clarified.
- Page 2 of the SOP describes a clean-up procedure for a spill. How does the laboratory determine that an area is “cleared” for resuming work after a spill?
- Do the samples entering the Luminex machine contain detergent (i.e. Tween) and confirm that this system is in fact a closed system.
- A3: BSL-3 should be changed to BSL-2*.
- Is there an appropriate disinfectant validated for inactivation of *Borrelia turicatae* and *Borrelia burgdorferi*?
- Medical surveillance issues are being discussed within OHS and an IBC sub-committee.
- Are there any documented cases of laboratory-acquired infections, and if so, how have they been handled?
- First Part C: C5a: Correct “HSV” to “*Borrelia turicatae*” and “stains” to “strains”.
- Second Part C: C5a: Correct “HSV” to “*Borrelia burgdorferi*”.

Dr. Crise made a motion to approve the registration, and recommended deferring the medical surveillance issues to a sub-committee, Dr. Garfinkel seconded and all were in favor. Dr. Baseler abstained from the vote.

07-34 (Dr. Hornung)

- Clarify procedures planned for BSL-2 work versus BSL 2* work.

Dr. Crise made a motion to approve, Mr. Winegar seconded and all were in favor. Dr. Baseler abstained from the vote.

07-35 (Dr. Hornung)

- Where will the blood be drawn (identify source material providers)?
- How is the blood transported to and from the lab?
- D9a: this was left blank
- C13a: Clarify where it is referenced that blood is aliquoted into nunc vials (is this for transport to next location?).

Dr. Hughes made a motion to approve with minor changes, Dr. Garfinkel seconded and all were in favor. Dr. Baseler abstained from the vote.

(06-106) (Merlino)

- A3: Why do all vectors used generate only defective nonproductive viruses? (Are there specific gene deletions that cause the vectors to be replication defective?)
- What potential hazards exist when the MLV-based vectors with oncogenes are put back into the mice? (i.e. Can the virus become mobilizable - mobilizable elements and unforeseen viruses in mice?)
- What would be the ramifications if an individual was stuck by a needle with a retroviral vector, which may or may not contains an oncogene?
- Explain how the presence of pBABE vector increases the hazards and provide a statement documenting awareness of the possibility of mobilization and recombination given the wide use of HIV-based vectors.
- B5a: Why is the virus that is made in the 293 packaging system nonproductive after infection into the target mammalian cell? (i.e. Are certain genes deleted that prevent replication?)
- B5g1: Since the cell lines are transfected with retroviral vectors, it is possible that there is presence of a human virus in the cell line before transfection. The retroviral vector could possibly recombine with that virus and become mobilizable. Why is this a possibility (what combinations of material may create this situation) and how will the risks associated with these potential hazards be addressed? What would be the ramifications of an individual if stuck by a needle with a retroviral vector which had been transfected into a human cell line?
- B6a: How do you know that once the cells are infected, they do not produce virus? (Is testing done, or is there documentation to support this?)
- D5a : Does the screening include human pathogen screening? The Laboratory of Molecular Technology performs a 10-panel screen for the presence of human pathogens in cell lines. This is required by the IBC when material will be introduced into animals.

- E6e1: Acknowledgement the recombination possibilities. The hazards associated with this possibility need to be identified and addressed.
- E6b1: States “we don’t do infections in the mouse”. How is this true if retroviral vectors and human cell lines are being introduced into the mouse?
- E9: If there is a possibility for recombination, what practices and procedures should be followed in the animal facility? Aerosols generated from cage changing and potential for viral shedding in animal excrement need to be mitigated. (i.e. mice should be housed in microisolator cages, sharps safety devices will be used, bedding will be dumped within a biological safety cabinet, etc.)
- Accordingly, animal care staff will need to be adequately informed and trained on the hazards associated with this work. This should be arranged with the animal facility manager.

Dr. Hughes made a motion to approve upon completion of the above requested modifications, Dr. Garfinkel seconded and all were in favor.

(06-94) Dimitrov

- A3: Provide more detailed information on the hazards associated with the potential for an occupationally transmitted HIV infection and how these hazards are mitigated.
- Clarify the use of Room 211, 211A, and 211B. Room 211 cannot have virus in it.
- Using the autoclave down the hallway is acceptable but be sure to transport waste in locking, sterilizable bins for hazardous waste.
- Clarify when and why things are taken out of the BSC when working with HIV? Does the material leave the room? If so, for what purpose? The material should not leave the BSL2* room.
- What are the cleaning and disinfecting procedures in place to avoid cross contamination issues? Does the p24 antigen assay use Tween to inactivate the material?
- Since the space will be shared with another group, what is the other group doing in the space so everyone using the space is familiar with the hazards present? There should be an acknowledgement between groups regarding the shared space that clearly defines work activities and hazards.

Dr. Baseler made a motion to conditionally approve this registration pending receipt of sufficient responses to the above issues, Dr. Hughes seconded and all were in favor.

(06-95) Dimitrov

- A1: Explain what will be done after expression takes place?
- Part B is blank. This will need to be completed since work is being conducted with recombinant vaccinia.
- C1a: Be more specific regarding the strain being used.
- Clarify homogenization versus sonification pertaining to the experiments.
- D3: should be “yes”
- D7: should be “yes”
- In the SOP - it should be noted that the work space is shared.
- Will vaccination be necessary for other room occupants? Evaluate their potential for exposure.

- D9a: needs more detail pertaining to safety operation measures.
- Describe the protein expression in vaccinia.
- A 1:10 solution of bleach is recommended for decontaminating materials that come into contact with the virus, making solutions at least weekly.
- Address the potential for occupational transmission and ocular hazards with respect to vaccinia.
- Explain how the phage and vaccinia will be kept separate
- Clean up procedures need to be more detailed, to include use of chemical disinfectants, concentrations, and how to address spills.

Dr. McVicar made a motion to defer approval, Dr. Crise seconded and all were in favor.

NEW BUSINESS

(07-30) Sterneck

- In the SOP, the use of safety glasses in the laboratory should be mandatory, not optional for this particular work.

Dr. Hughes made a motion to approve, Dr. Hollingshead seconded and all were in favor.

(07-31) Kalen

- Animal care personnel must be adequately informed of the hazards inherent to the protocol

Dr. Hollingshead made a motion to approve, Mr. Winegar seconded and all were in favor.

(07-36) McVicar

- No comments or suggestions were made regarding this registration.

Dr. Crise made a motion to approve, Dr. Hollingshead seconded and all were in favor.
Dr. McVicar abstained from the vote.

(07-27) Kaczmarczyk

- A1: The use of the 293 cells is unclear. Explain further what will be done with these cells.
- Differentiate between the use of GP2 cells and the 293 cells. These two systems need to be addressed separately.
- Do the 293 cells or the GP2 cells contain gag/pol?
- A3: Clarify the safety issues associated with the MMLV-VSVg.
- Provide a map of the reporter construct.

Dr. Crise made a motion to defer approval pending clarifications noted above, Mr. Winegar seconded and all were in favor.

AMENDMENTS

Sei (06-27)

- No comments or suggestions were made regarding this amendment.

Dr. Crise made a motion to approve the already modified amendment as is, Dr. Hughes seconded and all were in favor.

OTHER BUSINESS

- Dr. Charmaine Richman attended the meeting to request the committee to consider adding a question to the IBC Registration Form in an effort to remind Principal Investigators about the requirement to take the NIH “Protecting Human Subjects” on-line training. The question will be worded and sent to the committee for review. The question would be added to IBC form Part D question 11 or 2. A definition of human subjects will also be provided for clarification. This is a one-time training requirement.
- The Bloodborne Pathogen program is currently 96% compliant. There are now 1044 employees enrolled in the program.
- The IBC Charter was recently approved by the NCI and has been posted on the IBC webpage.

CONCLUSION

The meeting was adjourned at 2:00 p.m.

Theresa D. Bell, MPH, CBSP
IBC Secretary
Biological Safety Officer, EHS

Ms. Cara Leitch
IBC Coordinator
Sr. Safety Specialist, EHS

APPROVED:

Randall S. Morin, Dr. P.H.
Chairman, NCI-Frederick IBC
Director, EHS

Date

- xc: All Committee Members
Dr. Reynolds
Mr. Wheatley
Dr. Arthur
Mr. Bufter
Dr. Keimig