



Ebola cDNA: Biosafety Risks and Biosecurity Concerns

Biosafety Working Group Assessment

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Research with cDNA of Ebola (a RG4 Agent) in *E. coli*

- From a biosafety standpoint:
 - Is biosafety level 2 sufficient?
- From a biosecurity standpoint:
 - Full-length nucleic acid of Ebola in the proper setting can generate infectious virus—a Select Agent
 - However, full length RNA Ebola, by itself, is not considered to be a Select Agent nor is the cDNA
 - Are additional biosecurity measures warranted for this research beyond restrictions on access required for all research at BL2 containment (Appendix G-II-B-1-a)?

Is Biosafety Level 2 Sufficient?

- The complete cDNA from Ebola is not an irreversibly defective fraction of the genome; however,
 - Neither the full-length RNA genome nor the cDNA of Ebola is capable of producing infectious virus in prokaryotic or lower eukaryotic cells
 - Infectious Ebola virus can be rescued from its cDNA under very deliberate conditions in mammalian cells but can NOT be rescued from prokaryotic or lower eukaryotic cells

cDNA Manipulations of Ebola

- The full-length RNA genome of Ebola and the full length cDNA can produce infectious virus only with the addition of 4 essential viral proteins, (cDNA will also require an RNA polymerase)
- Single-strand negative-sense RNA viruses such as Ebola do not synthesize DNA – the cDNA copy is a laboratory construct

cDNA Manipulations of Ebola

- The rescue of infectious Ebola virus from its cDNA would require **scientific knowledge and appropriate skill**
- In addition, the rescue of infectious Ebola is a deliberate act
 - it can not be rescued accidentally

Working Group recommendations regarding Biosafety Containment

- Cloning of full length Ebola cDNA in *E. coli* K12 , a non-pathogenic prokaryote, can safely be performed at **BL2** due to the inherent biological properties of the cDNA in this system

Research Involving cDNA from Ebola in *E. coli*

From a biosecurity standpoint:

- Full-length nucleic acid of Ebola in the proper setting can generate infectious virus—a Select Agent
- However, full length Ebola RNA, by itself, is not currently considered to be a Select Agent nor is the cDNA

Standard Microbiological Practices (BL2)

Appendix G-II-B-1-a of the *NIH Guidelines* state:

“Access to the laboratory is limited or restricted by the Principal Investigator when work with organisms containing recombinant DNA molecules is in progress.”

- Are additional security measures warranted for this research beyond those specified above?

Addressing Biosecurity Concerns

- The *NIH Guidelines* are fundamentally about Biosafety not Biosecurity
- However, it is important to address the biosecurity implications of research with cDNA of Ebola
- What biosecurity measures are commensurate with the risk of generating an infectious Select Agent from Ebola cDNA?

CDC/NIH BMBL* on Biosecurity

- Definition of Biosecurity: protection of microbial agents from loss, theft, diversion or intentional misuse
- Biosafety and Biosecurity programs share common components:
 - Risk assessment
 - Personnel expertise and responsibility
 - Control and accountability for research materials including microorganisms and culture stocks
 - Access control elements
 - Material transfer documentation
 - Training, emergency planning and program management

Elements of a Biosecurity Program*

■ Program Management

- Organizational structure for the program and defines chain of command, roles and responsibilities

■ Physical Security

- Access to lab, freezers, etc. - limited to authorized and designated employees

■ Personnel Management

- Identify roles and responsibilities for employees who handle, use, store, and transport dangerous pathogens and/or other important assets
- Employee screening policies and procedures are used to evaluate the individuals who have access to pathogens

■ Inventory and Accountability

- Procedures for tracking inventory, storage, use, transfer and destruction of biological materials

Elements of a Biosecurity Program*

- Information security
 - Policies for handling sensitive information, e.g. security plans, access codes, agents inventories and storage locations
- Transport of Biological Agents
 - Material transport policies should include accountability measures for the movement of materials within an institution and outside the facility
- Accident, Injury and Response Plans
- Chain of Notification established in advance of an actual event
- Training and Practice Drills
- Security Updates and Reevaluations

RML-Proposed Biosecurity Measures for Cloning the full-length cDNA of Ebola virus in *E. coli* at BL2

- Access to laboratory limited by separate secure space (a lab within a lab)
- Material accountability procedures to be implemented to track inventory, storage, use, transfer and destruction of dangerous biological materials
- Personnel required to have Public Trust Level 5 Clearance* and appropriate training

* Public Trust Level is only available to Federal employees and contractors

WG recommendation: Biosecurity Procedures for Research with cDNA in *E. coli* at BL2

- Re-evaluate proposed security measures for this research using the BMBL Principles of Biosecurity and establish a security plan that includes the proposed measures and any additional necessary elements including:
 - Appointment of an appropriate official to oversee the security plan on behalf of the Institution
 - Establishment of a training program for the security aspects of the research in addition to biosafety
 - Re-evaluate the program periodically and update as needed

WG Recommendations: cDNA of Ebola into *E. coli*

- **Biosafety Risk Assessment:** Cloning of the full cDNA of Ebola into *E. coli* can be safely done at BL2
- **Biosecurity:** In keeping with the approach outlined in the BMBL, the PI and Institution is responsible for developing and implementing an appropriate biosecurity plan that addresses the principles of biosecurity outlined in the BMBL and is commensurate with the risk.
- WG recommendation to work with the cDNA of Ebola at BL2 in the non-pathogenic prokaryote *E. coli* K12 is specific to Dr. Feldmann at RML

WG Recommendations for Future Requests Regarding cDNA of Ebola into *E. coli*

- OBA will review future requests to do the same type of research at BL2 containment on a case-by-case basis
- OBA will conduct, in consultation with the RAC as needed, a biosafety risk assessment to determine if such research can be safely conducted at BL2

WG Recommendations for Future Requests Regarding cDNA of Ebola into *E. coli*

- In keeping with the approach outlined in the Section VI of the BMBL, Principles of Biosecurity, it is the responsibility of the Institution to perform a biosecurity risk assessment and develop an appropriate plan
 - Applications should therefore include 1) a description of the biosafety risk assessment undertaken in accordance with the *NIH Guidelines* and 2) the biosecurity assessment and plan using Section VI of the BMBL 5th edition.