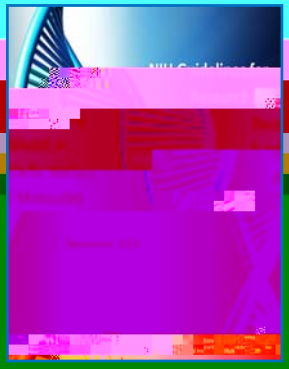
# Overview of the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules*

National Institutes of Health
Office of Biotechnology Activities

## *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules*

**A scientifically-responsive document that will continue to evolve**



**Has undergone multiple revisions since 1976**

**Latest version – November 2013**

**http://osp.od.nih.gov/office-biotechnology-activities/biosafety/nih-guidelines**

## Content of the *NIH Guidelines*

Section I – Scope

Section II – Safety Considerations

Section III – Types of Experiments Covered

Section IV – Roles and Responsibilities

Appendices

## *NIH Guidelines* – Section I

### Scope and Applicability

Specifies practices for constructing and handling

(i) recombinant nucleic acid molecules,

(ii) synthetic nucleic acid molecules, including those that are chemically or otherwise modified but can base pair with naturally occurring nucleic acid molecules, and

(iii) cells, organisms and viruses containing such molecules.

## NIH Guidelines – Section I

- In the context of the NIH Guidelines, recombinant and synthetic nucleic acids are defined as:
  - (i) molecules that a) are constructed by joining nucleic acid molecules and b) can replicate in a living cell, i.e. recombinant nucleic acids;
  - (ii) nucleic acid molecules that are chemically or by other means synthesized or amplified, including those that are chemically or otherwise modified but can base pair with naturally occurring nucleic acid molecules, i.e. synthetic nucleic acids; or
  - (iii) molecules that result from the replication of those described in (i) or (ii) above.

## The NIH Guidelines Apply to…

- Research with recombinant or synthetic (or both) nucleic acid molecules that is
  - Performed at or sponsored by an institution that receives any NIH funding for such research
- Rationale: For biosafety to be meaningful, it has to be observed by all investigators at an institution

## Are the *NIH Guidelines* Optional?

**"Guidelines" does not mean "optional"**

**They are a term and condition of NIH funding for research with recombinant or synthetic nucleic acid molecules**

## Are the *NIH Guidelines* Optional?

**What are potential consequences of noncompliance with the *NIH Guidelines*?**

**Suspension, limitation, or termination of NIH funds for research subject to the *NIH Guidelines* at the institution, or**

**A requirement for prior NIH approval of any or all research subject to the *NIH Guidelines* at the institution.**

## Prescription versus Flexibility

- Some matters are left to institutional discretion
- Flexibility is a two-sided coin
  - Accommodates institutional diversity and heterogeneity
  - Can create uncertainty about expectations

## Specifics vs. Intent

"The *NIH Guidelines* will never be complete or final since all conceivable experiments involving recombinant or synthetic nucleic acid molecules cannot be foreseen. Therefore, it is the responsibility of the institution and those associated with it to adhere to the intent of the *NIH Guidelines* as well as to the specifics."

- Good judgment is key
- OBA can help

## Section II - Safety Considerations

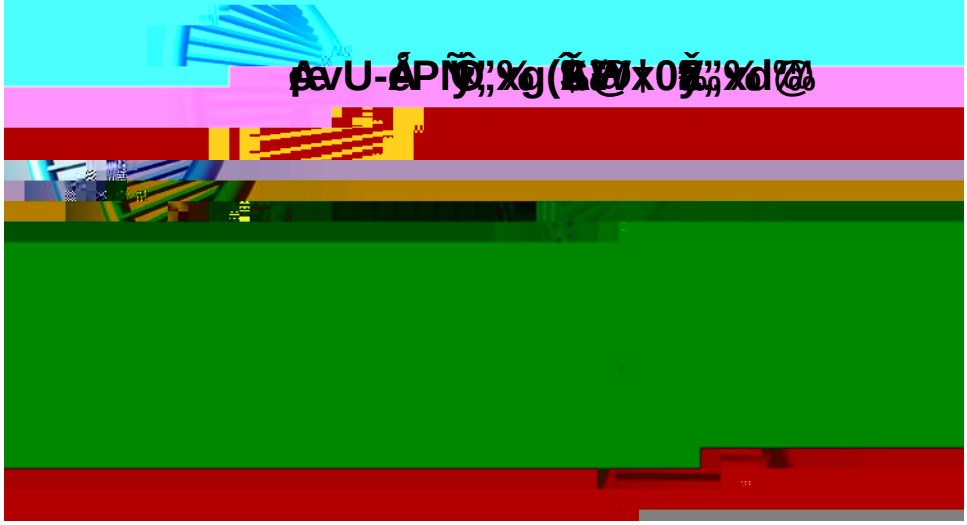
## *NIH Guidelines* – Section II

### Safety Considerations

**Risk assessments: (Appendix B)**

| RG 1 | RG 2 | RG 3 | RG 4 |
|---|---|---|---|
| Agents that are not associated with disease in healthy adult humans | Agents that are | | |

# *NIH Guidelines* – Section II



## Section III-A

## Section III-B

**Experiments Require NIH/OBA and IBC Approval Before Initiation**

**III-B-1: Experiments involving the cloning of toxin molecules with**

**III-B-2: Experiments that have been approved (under Sec**

# Section III-C

**Experiments Require RAC Review, IBC Approval and IRB Approval <u>Before</u> Initiation**

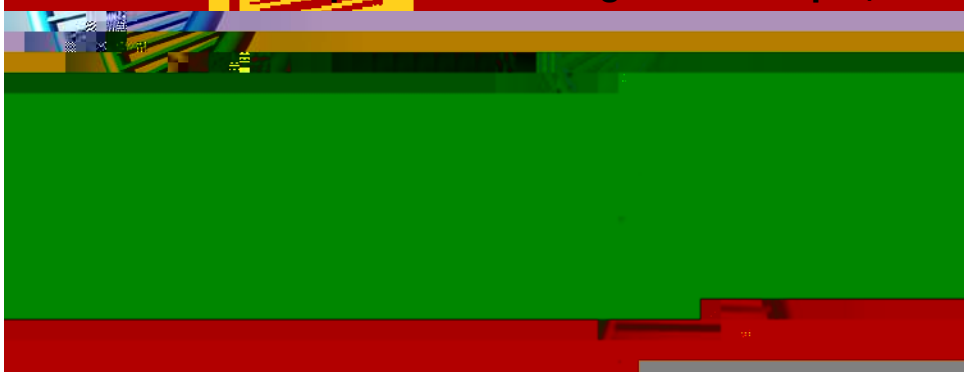
**Human gene transfer - deliberate transfer into human research participants of either:**

- **Recombinant nucleic acid molecules, or DNA or RNA derived from recombinant nucleic acid molecules, or**
- **Synthetic nucleic acid molecules, or DNA or RNA derived from synthetic**

# Section III-D-1

## Experiments IBC Require Approval Before Initiation

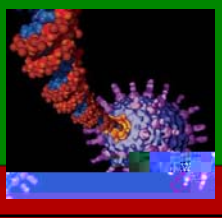
**Experiments Using Risk Group 2,**

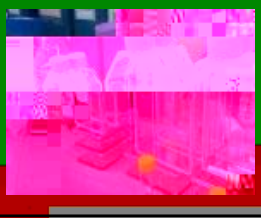


# Section III-D-3

## Experiments Require IBC Approval Before Initiation

Experiments Involving the Use of Infectious DNA or RNA Viruses or Defective DNA or RNA Viruses in the Presence of Helper Virus in Tissue Culture Systems



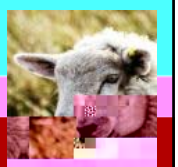


# Section III-D-4: Experiments Involving Whole Animals

## Includes experiments in which:

The animal's genome has been altered by stable introduction of recombinant or synthetic nucleic acids into germline (transgenic animals)

Viable recombinant or synthetic nucleic acid molecule-modified microorganisms are tested on whole animals



# Section III-D-7

## Experiments Involving Influenza Viruses

## Section III-E-3

### Experiments Involving the Generation of Transgenic Rodents

**Experiments in which:**

- Rodent's genome has been altered by stable introduction of recombinant or synthetic nucleic acid molecules into germline
- BL1 containment is appropriate

## Section III-F: Exempt Experiments

**Registration with the Institutional Biosafety Committee is not required (although many institutions may require this by policy)**

## Section III-F-1: Exempt Experiments

### Synthetic nucleic acids that:

(1) can neither replicate nor generate nucleic acids that can replicate in any living cell (e.g., oligonucleotides or other synthetic nucleic acids that do not contain an origin of replication or contain elements known to interact with either DNA or RNA polymerase), and

(2) are not designed to integrate into DNA, and

(3) do not produce a toxin that is lethal for vertebrates at an LD50 of less than 100 nanograms per kilogram body weight.

## Section III-F-1: Exempt Experiments

**Note:** If a synthetic nucleic acid is deliberately transferred into one or more human research participants and meets the amended criteria of Section III-C, it is not exempt under the *NIH Guidelines*.

# Section III-F-2

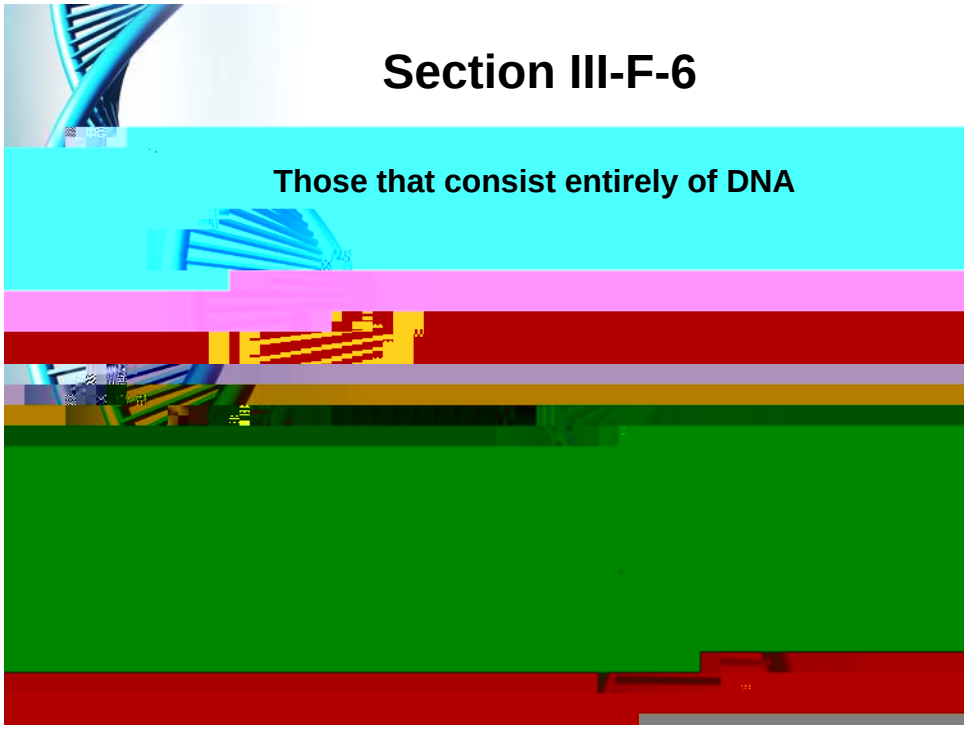
Those that are not in organisms, cells or viruses and that have not been modified or manipulated (e.g., encapsulated into

## Section III-F-4

Those that consist entirely of nucleic acids from a prokaryotic host including its indigenous plasmids or viruses when propagated only in that host (or a closely related strain of the same species), or when transferred to another host by well established physiological means.

## Section III-F-5

Those that consist entirely of nucleic acids from an eukaryotic host including its chloroplasts, mitochondria, or plasmids (but excluding viruses) when propagated only in that host (or a closely related strain of the same species).


Section III-F-6
Those that consist entirely of DNA

## Section III-F-8

**Those that do not present a significant risk to health or the environment as determined by the NIH Director, with the advice of the RAC, and following appropriate notice and opportunity for public comment.**

**See Appendix C, *Exemptions under***

# Appendix C-II

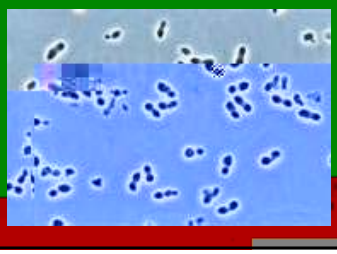
## *Escherichia coli K-12* Host-Vector Systems

**Experiments which use *Escherichia coli* K-12 host-vector systems (with the exception of those**

## Appendix C-IV

### *Kluyveromyces* Host-Vector Systems

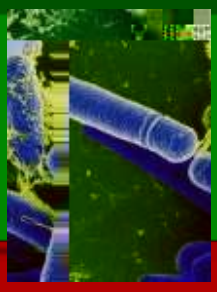
Experiments involving K. lactis host-vector systems (with the exception of experiments listed in Appendix C-III-A) are exempt



## Appendix C-V

### *Bacillus subtilis* or *Bacillus licheniformis* Host-Vector Systems

Any asporogenic *Bacillus subtilis* or asporogenic *Bacillus licheniformis* strain which does not revert to a spore-former with a frequency greater than $10^7$ may be used for cloning DNA (with the exception of those experiments listed in Appendix C-IV-A, *Exceptions*)



# Appendix C-VI

## Extrachromosomal Elements of Gram Positive Organisms

**Recombinant or synthetic nucleic acid molecules derived entirely from extrachromosomal elements of the organisms listed below, propagated and maintained in organisms listed below are exempt.**

***Bacillus amyloliquefaciens***
***Bacillus amylosaccharitícus***
***Bacillus anthracis***
***Bacillus aterrimus***
***Bacillus brevis***
***Bacillus cereus***
***Bacillus globigii***
***Bacillus licheniformis***
***Bacillus megaterium…….*** **(see *NIH Guidelines* for complete list)**

# Appendix C-VII

## The Purchase or Transfer of Transgenic Rodents

**The purchase or transfer of transgenic rodents for experiments that require IBC**

# Appendix C-VIII

## Generation of BL1 Transgenic Rodents via Breeding

**The breeding of two different transgenic rodents or the breeding of a transgenic rodent and a non-transgenic rodent with the intent of creating a new strain of transgenic rodent that can be housed at BL1 containment will be exempt from the *NIH Guidelines* if:**

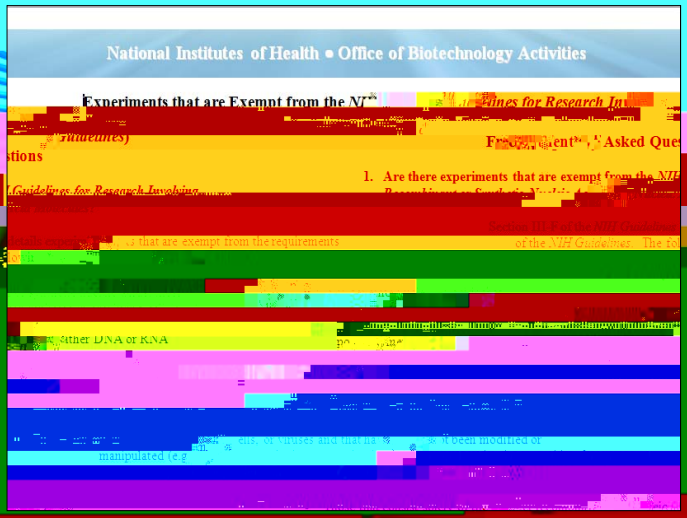
**(1) Both parental rodents can be housed under BL1 containment; and**

**(2) neither parental transgenic rodent contains the following genetic modifications: (i) incorporation of more than one-half of the genome of an exogenous eukaryotic virus from a single family of viruses; or**

**(ii) incorporation of a transgene that is under the control of a gammaretroviral long terminal repeat (LTR); and**

**(3) the transgenic rodent that results from this breeding is not expected to contain more than one-half of an exogenous viral genome from a single family of viruses.**

# Section III-F (and Appendix C)



## NIH Guidelines – Section IV

### Roles and Responsibilities

- Institution
- Institutional Biosafety Committee (IBC)
- Biological Safety Officer (BSO)
- Principal Investigator (PI)
- NIH

## Institutional Responsibilities under the *NIH Guidelines*

**The Institution shall:**

- Establish and implement policies for the safe conduct of research subject to the *NIH Guidelines*
- Establish an Institutional Biosafety Committee
- Assist and ensure compliance with the *NIH Guidelines* by investigators
- Ensure appropriate training for IBC members and staff, PIs, laboratory staff
- Determine necessity for health surveillance of personnel
- Report any significant accidents, incidents or violations to OBA within 30 days (or immediately as required)

# PI Responsibilities under the *NIH Guidelines*

**The Principal Investigator shall (among other things):**

- **Initiate or modify no research subject to the *NIH Guidelines* which requires IBC approval until approval is granted**
- **Determine whether experiments are covered under III-E and notify the IBC as appropriate**
- **Be adequately trained in good microbiological techniques**
- **Adhere to IBC emergency plans for spills and personnel contamination**
- **Report any significant problems or violations to OBA within 30 days (or immediately as required)**

## NIH OBA Responsibilities under the *NIH Guidelines*

### Basic experiments reviewed by NIH OBA

- Deliberate transfer of drug resistance trait to microorganisms not known to acquire the trait naturally, if it could compromise disease control
- Cloning of toxin molecules with $LD_{50}$ <100 ng/Kg bodyweight
- Recombinant or synthetic nucleic acid molecules from restricted agents transferred to nonpathogenic prokaryotes or lower eukaryotes
- Recombinant or synthetic nucleic acid molecules from nonpathogenic prokaryotes or lower eukaryotes transferred to restricted agents
- Use of infectious or defective restricted poxviruses in presence of helper virus

## *NIH Guidelines* - Appendices

| | |
|---|---|
| Appendix A – | Exemptions: Natural Exchangers |
| Appendix B – | Classification of Etiologic Agents |
| Appendix C – | Exemptions under III-F |
| Appendix D – | Major Actions |
| Appendix E – | Certified Host-Vector Systems |
| Appendix F – | Biosynthesis of Toxic Molecules |
| Appendix G – | Physical Containment |

# Organization of the *NIH Guidelines*

**Appendix J –** **Biotechnology Research Subcommittee**

**Appendix K –** **Large Scale Physical**