

The Guidelines Explained: Recombinant DNA research

Source of documentation: Harvard Institutional Biosafety Committee

This document was prepared to capture the essence of the NIH guideline requirements and put them into readable form. Many details are omitted, the actual guidelines should be consulted when in doubt. The NIH recombinant DNA guidelines are an intimidating publication. They are long, full of legalese, and have lots of cross references, exceptions, lists, sections, appendices and tables. The NIH rDNA guidelines are legally binding on any institution using NIH funds.

Registration

The NIH requires all labs working with recombinant DNA to register the work with their local Institutional Biosafety Committee (IBC). The guidelines distinguish among five kinds of registrations. The type depends on the potential hazard of the work. More hazardous means more approvals are needed. The five types are:

- Work that cannot begin until there is NIH and IBC approval (Sections IIIA- and III-B) this is the most dangerous level and it is **[Taboo]** (page 2)
- Work that cannot begin until there is a RAC review and then IBC approval (Section III-C). This tends to involve sensitive and potentially dangerous agents **[Wait and Wait]** (page 2).
- All other recombinant DNA work cannot begin until there is IBC approval (Section III-D), there is usually a short **[Wait]** (page 2). Human Xenotransplantation Studies are in this category (this requirement comes from the FDA).
- The NIH Guidelines describe two other classes: those that require IBC notification (Section III-E) and those that are completely exempt from NIH requirements (Section III-F)

What is recombinant DNA?

It is either a) DNA constructed *in vitro* from separate DNA segments that can replicate and/or express a biologically active polynucleotide or polypeptide *in vivo*, or b) synthetic DNA that has the potential of generating a hazardous product *in vivo*.

What are the RAC and the OBA?

The NIH OBA (Office of Biotechnology Activities) is an administrative arm responsible for carrying out the orders of the NIH Director with regard to recombinant DNA, genetic testing and xenotransplantation. An advisory committee is involved in establishing policies for each of these fields.

‘Taboo’

*(NIH approval and IBC permission required)
From NIH Guidelines Sections IIIA and IIIB*

- ❖ Making drug resistant constructs of microorganisms if they compromise the drug’s therapeutic potential,
- ❖ Making constructs that synthesize vertebrate toxins with an LD₅₀ of 100ng/mg and 100µg/mg

‘Wait and Wait’

*(NIH Review and IBC permission required)
From NIH Guidelines Section III-C*

- ❖ Studies in which genes are transferred into human must be submitted to the NIH OBA for review. If the OBA finds the study to be “novel” it will place the study on the next RAC meeting agenda. IBC approval must wait for the RAC’s review. If, on the other hand, OBA does not deem the study to be “novel”, IBC can act immediately.

‘Wait’

*(IBC Review Needed Before Starting)
From Section III-D and III-E*

To date, all studies carried out at USA are in this group. They are examined by the IBC with an eye to recommending safe procedures and containment. To reach a conclusion it is often useful to classify the risk associated with a proposed study according to the risk associated with the organisms to be used. A convenient classification tool is the concept of “risk group.”

Risk Groups

The NIH classifies biological agents into four risk groups according to their human pathogenicity (NIH guidelines, Section II-A-1)

- ❖ Risk Group 1: not associated with disease in healthy adults
- ❖ Risk Group 2: associated with disease that is rarely serious and for which therapeutic or preventive options are often available.
- ❖ Risk Group 3 : associated with serious or lethal disease for which therapeutic or preventive options may be available.
- ❖ Risk Group 4: associated with serious or lethal disease for which therapeutic or preventive options not usually available.

** Appendix B in the NIH Guidelines list biological agents according to their Risk Group **

In general, the Risk Group determines the Biosafety level needed: for instance a Risk Group 3 is usually studied in a BL-3 lab.

‘Low-Hazard’ Recombinant DNA (Section III-E)

- [Non-pathogenic prokaryotes or non-pathogenic lower eukaryotes (use BSL-1)
- [Recombinant DNA with less than 2/3 of a eukaryotic viral genome (and no helper) used exclusively in tissue culture (BL1 recommended)
- [Some hosts *Plants* carrying recombinant DNA. BL1 containment is recommended when the host is:
 - A non-noxious weed
 - A plant or microorganism thought not to damage the ecosystem

- [Some other host *Plants* carrying recombinant DNA. BL2 or BL1 plus level containment is recommended when:
- The host is a noxious weed
 - The introduced DNA is the complete genome of an infectious agent known to normally exist in the US,
 - The host is a plant or microorganism that may damage ecosystems,
 - The host is a plant with a recombinant DNA from foreign microorganisms that are thought to be safe for the ecosystem

‘No Waiting’

(Just notify)

From Section III-D

Exempt Recombinant DNA – (Section III-F and Appendix C)

“Exempt” from NIH guidelines means that work with these constructs need to be submitted to the IBC for notification. The IBC will forward a exemption letter for the rDNA being used in the research. It is USA policy for all registered “exempt” DNA recombinant research to be conducted at BSL-1.

Exempt experiments include:

- Those that are not in organisms or viruses;
- Those that consist entirely of DNA segments from a single nonchromosomal or viral DNA source, though one or more of the segments may be a synthetic equivalent;
- Those that consist entirely of DNA 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;
- Those that consist entirely of DNA from an eukaryotic host including its chloroplasts, mitochondria, or plasmids (but excluding viruses) when propagated only in the host (or a closely related strain of the same species);
- Those that consist entirely of DNA segments from different species that exchange DNA by known physiological processes, though one or more of the segments may be a synthetic equivalent. See Appendices A-I through A-VI, Exemptions Under Section III-F-5 – Sublists of Natural Exchangers, for a list of natural exchangers that are exempt from the NIH Guidelines; or
- Those that do not present a significant risk to health or the environment (*see section IV-C-1-b-(1)-(c), Major Actions*)