

IBC  
APPENDIX R  
ANIMAL BIOSAFETY LEVEL (ABSL) DETERMINATION MATRIX

A guideline based on the current edition of the *Biosafety in Microbiological and Biomedical Laboratories (BMBL)\** which is to be used for determining BSL and ABSLs for research  
(\* [https://www.cdc.gov/labs/media/pdfs/2025/08/SF\\_19a\\_308133-A\\_BMBL6\\_00-BOOK-WEB-final-3.pdf](https://www.cdc.gov/labs/media/pdfs/2025/08/SF_19a_308133-A_BMBL6_00-BOOK-WEB-final-3.pdf))

| NOTE: The following PPE must be worn in all facilities when entering animal rooms or labs where animals are manipulated: <i>facility dedicated scrubs/shoes, gloves, surgical mask, eye protection (or face shield). Additionally, all cage dump stations must be ventilated.</i> |  |   |                               |   |   |   |  |
|---|--|---|-------------------------------|---|---|---|--|
| MATERIAL  | BIOSAFETY LEVEL (applicable to injections/necropsy)  | ANIMAL BIOSAFETY LEVEL (applicable for animal housing / cage changes) | HOUSING REQUIREMENTS          | CAGE CHANGES  | PRE/POST INFECTION MANIPULATIONS  | IMAGING   |  |
| <b>MATERIALS NOT KNOWN TO BE TRANSMITTED VIA AEROSOL ROUTES (* "Retroviral" is inclusive of lentiviral vectors)</b>   |  |   |                               |   |   |   |  |
| 1   | Murine cell lines transduced with an ecotropic retroviral* vector with benign insert (ex. GFP)       | BSL2  | ABSL1                         | No special requirements   | <b>No special requirements</b> - cages can be changed on benchtop. Bedding does not need to be autoclaved prior to disposal / cages do not need to be autoclaved prior to washing | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | No special requirements  |
| 2   | Murine cell lines transduced with an amphotropic retroviral* vector with benign insert (ex. GFP)     | BSL2  | ABSL2 for 1 week / then ABSL1 | Microisolator technique for 1 week post dosing; after 1 week, no special requirements | <b>For first week:</b> 1. ALL cage changes in BSC; 2. bag and autoclave bedding; 3. bag and autoclave caging before wash; After one week, resume normal cage change.              | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | All manipulations inside BSC where feasible. Maintain BSL2/ABSL2 practices where feasible, especially during the first week. |
| 3   | Murine cell lines transduced with an ecotropic retroviral* vector with 'hot' insert (ex. oncogene)   | BSL2  | ABSL1                         | No special requirements   | <b>No special requirements</b> - cages can be changed on benchtop. Bedding does not need to be autoclaved prior to disposal / cages do not need to be autoclaved prior to washing | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | No special requirements  |
| 4   | Murine cell lines transduced with an amphotropic retroviral* vector with 'hot' insert (ex. oncogene) | BSL2  | ABSL2 for 1 week / then ABSL1 | Microisolator technique for 1 week post dosing; after 1 week, no special requirements | <b>For first week:</b> 1. ALL cage changes in BSC; 2. bag and autoclave bedding; 3. bag and autoclave caging before wash; After one week, resume normal cage change.              | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | All manipulations inside BSC where feasible. Maintain BSL2/ABSL2 practices where feasible, especially during the first week. |

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|--|---|--|--|---|---|---|--|
|  | MATERIAL  | BIOSAFETY LEVEL<br>(applicable to injections/necropsy) | ANIMAL BIOSAFETY LEVEL<br>(applicable for animal housing / cage changes) | HOUSING REQUIREMENTS  | CAGE CHANGES  | PRE/POST INFECTION MANIPULATIONS  | IMAGING  |
| 5  | Murine cell lines transduced with an pantropic retroviral* vector with 'hot' insert (ex. oncogene)  | BSL2   | ABSL2 for 1 week / then ABSL1  | Microisolator technique for 1 week post dosing; after 1 week, no special requirements | <b>For first week:</b> 1. ALL cage changes in BSC; 2. bag and autoclave bedding; 3. bag and autoclave caging before wash; After one week, no special requirements                 | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | All manipulations inside BSC where feasible. Maintain BSL2/ABSL2 practices where feasible, especially during the first week. |
| 6  | Unmodified murine cells into mice   | BSL1   | ABSL1  | No special requirements   | <b>No special requirements</b> - cages can be changed on benchtop. Bedding does not need to be autoclaved prior to disposal / cages do not need to be autoclaved prior to washing | No special requirements   | No special requirements  |
| <b>MATERIALS KNOWN TO BE TRANSMITTED VIA AEROSOL ROUTES</b>  |   |  |  |   |   |   |  |
| 7  | Adenovirus/Adeno-Cre; Adenoviral vectors; cells transduced with adenoviral vectors  | BSL2   | ABSL2 for 1 week / then ABSL1  | Microisolator technique for 1 week post dosing; after 1 week, no special requirements | <b>For first week:</b> 1. ALL cage changes in BSC; 2. bag and autoclave bedding; 3. bag and autoclave caging before wash; After one week, resume normal cage change.              | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | All manipulations inside BSC where feasible. Maintain BSL2/ABSL2 practices where feasible, especially during the first week. |
| 8  | Known Human Pathogens (ex. <i>Toxoplasma gondii</i> ; <i>Listeria monocytogenes</i> ; <i>Helicobacter pylori</i> ; <i>LCMV</i> ; <i>Citrobacter rodentium</i> ; Certain toxins (i.e. Diphtheria); others as reviewed by IBC | BSL2   | ABSL2  | Microisolator technique for DURATION of experiment                                    | <b>For first week:</b> 1. ALL cage changes in BSC; 2. bag and autoclave bedding; 3. bag and autoclave caging before wash; After one week, resume normal cage change.              | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | All manipulations inside BSC where feasible. Maintain BSL2/ABSL2 practices where feasible.                                   |

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| NOTE: The following PPE must be worn in all facilities when entering animal rooms or labs where animals are manipulated: <i>facility dedicated scrubs/shoes, gloves, surgical mask, eye protection (or face shield). Additionally, all cage dump stations must be ventilated.</i> |  |  |                               |   |  |   |  |
|---|--|--|-------------------------------|---|--|---|--|
| MATERIAL  | BIOSAFETY LEVEL<br>(applicable to injections/necropsy)   | ANIMAL BIOSAFETY LEVEL<br>(applicable for animal housing / cage changes) | HOUSING REQUIREMENTS          | CAGE CHANGES  | PRE/POST INFECTION MANIPULATIONS   | IMAGING   |  |
| <b>HUMAN CELL LINES (* "Retroviral" is inclusive of lentiviral vectors)</b>   |  |  |                               |   |  |   |  |
| 9   | Unmodified (established) Human cell lines inserted into an Immuno-compromised mouse                | BSL2   | ABSL1                         | No special requirements   | No special requirements - cages can be changed on benchtop. Bedding does not need to be autoclaved prior to disposal / cages do not need to be autoclaved prior to washing | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | No special requirements  |
| 10  | Human cell lines transduced with retroviral* vector then inserted into an immuno-compromised mouse | BSL2   | ABSL2 for 1 week / then ABSL1 | Microisolator technique for 1 week post-dosing; after 1 week, no special requirements | <b>For first week:</b> 1. ALL cage changes in BSC; 2. bag and autoclave bedding; 3. bag and autoclave caging before wash; After one week, resume normal cage change.       | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | All manipulations inside BSC where feasible. Maintain BSL2/ABSL2 practices where feasible, especially during the first week. |
| 11  | PRIMARY human tissue transplant into mice  | BSL2   | ABSL2                         | Microisolator technique for DURATION of experiment                                    | 1. ALL cage changes in BSC; 2. bag and autoclave bedding; 3. bag and autoclave caging before wash  | All injections and necropsies in BSC for DURATION of project (following BSL2 practices) | All manipulations inside BSC where feasible. Maintain BSL2/ABSL2 practices where feasible.                                   |