

Routine Health Monitoring of Charles River Mice and Rat Barrier Production Colonies in North America

Health Monitoring Testing:

This document describes the routine health monitoring of immunocompetent mice and rats raised in Charles River barrier production colonies. These rodents are referred to as VAF/Plus[®], which indicates that the animals have been tested and are free of an extensive list of viruses and other pathogens. Please note that only immunocompetent animals are raised in these colonies. Immunodeficient models and certain immunocompetent animals, such as the VAF Elite[®] mice and rats, are raised using different housing, husbandry and biosecurity techniques, and health monitoring programs for those animals are described separately.

- Every four weeks, Charles River uses the exquisitely sensitive TaqMan[®] PCR to gain a snapshot of whole-room health status. Ten environmental samples are collected from a variety of sites, including air exhaust grates and bedding disposal equipment. This PCR-based testing includes common agents whose detection would trigger immediate cessation of shipping.
- Every four weeks, at the two-week point between PCR tests, Charles River performs serologic monitoring using the sensitive Multiplexed Fluorometric ImmunoAssay[®] on 16 animals from each mouse and rat production room. In barrier production rooms with multiple strains of animals, the 16 in the test set includes eight from two different strains.

- Every 13 weeks (quarterly), Charles River evaluates 12–16 animals at three different age groups from each barrier room. Necropsy samples are evaluated by direct parasitology examination, microbiological cultures, gross pathology and limited PCR testing.
- Annually, Charles River uses an innovative PCR rodent infectious agent (PRIA) panel to evaluate 12–16 animals at three different age groups from each barrier room for an extensive list of viruses, bacteria, fungi and parasites.

Table 1 at the end of this document summarizes agents tested and the methods used to monitor all Charles River mouse and rat barrier production colonies. Charles River is also capable of performing alternate procedures to enable confirmatory testing in order to corroborate unexpected positive results. The complete Charles River health monitoring program is regularly reviewed and revised as needed to ensure that Charles River colonies are evaluated in a scientifically rigorous, valid and cost-effective manner.

Communication:

Charles River communicates the health status of the barrier production colonies primarily through the Charles River website, where colony health status reports are updated weekly to include any new results. Shipments of animals in some regions may also include colony status results from the time the animals were packed. In addition to the pathogens included on the VAF/Plus® lists whose confirmed presence would result in immediate colony termination, Charles River also reports surveillance results for some agents that are typically considered to be of little consequence in immunocompetent animals, primarily human commensal bacteria. These agents are reported as a service to clients whose specialized areas of research may require

consideration of additional microorganisms. Colony health reports include a description of which action would be taken for each listed agent.

It remains Charles River's policy to inform customers in a timely manner of any breaches in animal health or genetic integrity, providing urgent colony health information via email or other method. For assistance regarding specific information on Charles River monitoring procedures, additional data on animals, or interpretation of the monitoring information, please direct inquiries to Charles River Technical Services (1-877-274-8371) or email askcharlesriver@crl.com.

Table 1: Summary of Methods Used to Screen Charles River North American Immunocompetent Mouse and Rat Colonies

| Agent/Virology | Sampling Site or Method | Rat Testing Frequency (Weeks) | Mouse Testing Frequency (Weeks) |
|---------------------------------------------|-------------------------|-------------------------------|---------------------------------|
| Ectromelia Virus (ECTRO) | Serum | - | 13 |
| Hantaan Virus (HANT) | Serum | 13 | 13 |
| Kilham Rat Virus (KRV) | Serum | 4 | - |
| Lactate Dehydrogenase-Elevating Virus (LDV) | Serum | - | 13 |
| Lymphocytic Choriomeningitis Virus (LCMV) | Serum | 13 | 13 |
| Minute Virus of Mice (MVM) | Serum | - | 4 |
| Mouse Adenovirus (MAV1&2) | Serum | 13 | 13 |
| Mouse Cytomegalovirus (MCMV) | Serum | - | 13 |
| Mouse Hepatitis Virus (MHV) | Serum | - | 4 |
| Mouse Parvovirus (MPV) | Serum | - | 4 |
| Mouse Pneumotropic Virus (K) | Serum | - | 13 |
| Mouse Rotavirus (EDIM) | Serum | - | 4 |
| Mouse Thymic Virus (MTLV) | Serum | - | 13 |
| Murine Norovirus (MNV) | Serum | - | 4 |

Table 1: continued

| Agent/Virology | Sampling Site or Method | Rat Testing Frequency (Weeks) | Mouse Testing Frequency (Weeks) |
|-------------------------------------------------------|-------------------------|-------------------------------|---------------------------------|
| Pneumonia Virus of Mice (PVM) | Serum | 4 | 13 |
| Polyoma Virus (POLY) | Serum | - | 13 |
| Rat Coronavirus (RCV/SDAV) | Serum | 4 | - |
| Rat Minute Virus (RMV) | Serum | 4 | - |
| Rat Parvovirus (RPV) | Serum | 4 | - |
| Rat Theilovirus (RTV) | Serum | 4 | - |
| Reovirus (REO) | Serum | 4 | 13 |
| Sendai Virus (SEND) | Serum | 4 | 13 |
| Theiler's Murine Encephalomyelitis Virus (TMEV-GDVII) | Serum | - | 4 |
| Toolan's H-1 Virus (H1) | Serum | 4 | - |
| Agent/Microbiology | Sampling Site or Method | Rat Testing Frequency (Weeks) | Mouse Testing Frequency (Weeks) |
| Beta hemolytic <i>Streptococcus</i> sp. | Respiratory tract | 13 | 13 |
| Beta hemolytic <i>Streptococcus</i> sp. –Group B | Respiratory tract | 13 | 13 |
| Beta hemolytic <i>Streptococcus</i> sp. –Group G | Respiratory tract | 13 | 13 |
| <i>Bordetella bronchiseptica</i> | Respiratory tract | 13 | 13 |
| CAR bacillus | Serum | 13 | 13 |
| <i>Citrobacter rodentium</i> | Gastrointestinal tract | - | 13 |
| <i>Corynebacterium kutscheri</i> | Respiratory tract | 13 | 13 |
| <i>Helicobacter</i> sp. | Fecal pellets | 13 | 13 |
| <i>Helicobacter bilis</i> | Fecal pellets | 13 | 13 |
| <i>Helicobacter hepaticus</i> | Fecal pellets | 13 | 13 |

Table 1: continued

| Agent/Microbiology | Sampling Site or Method | Rat Testing Frequency (Weeks) | Mouse Testing Frequency (Weeks) |
|----------------------------------------|----------------------------|-------------------------------|---------------------------------|
| <i>Klebsiella oxytoca</i> | Respiratory tract | 13 | 13 |
| | Gastrointestinal tract | 13 | 13 |
| <i>Klebsiella pneumoniae</i> | Respiratory tract | 13 | 13 |
| | Gastrointestinal tract | 13 | 13 |
| <i>Mycoplasma pulmonis</i> | Serum | 4 | 13 |
| <i>Pasteurella pneumotropica</i> | Respiratory tract | 13 | 13 |
| <i>Pasteurella multocida</i> | Respiratory tract | 13 | 13 |
| <i>Pneumocystis carinii</i> | Serum | 4 | - |
| <i>Pseudomonas aeruginosa</i> | Respiratory tract | 13 | 13 |
| | Gastrointestinal tract | 13 | 13 |
| <i>Salmonella</i> spp. | Gastrointestinal tract | 13 | 13 |
| <i>Staphylococcus aureus</i> | Respiratory tract | 13 | 13 |
| <i>Streptobacillus moniliformis</i> | Respiratory tract | 52 | 52 |
| <i>Streptococcus pneumoniae</i> | Respiratory tract | 13 | 13 |
| Tyzer's Disease | Clinical and/or gross exam | 13 | 13 |
| Agent/Parasitology | Sampling Site or Method | Rat Testing Frequency (Weeks) | Mouse Testing Frequency (Weeks) |
| Ectoparasites | Direct exam | 13 | 13 |
| <i>Encephalitozoon cuniculi</i> (ECUN) | Serum | 13 | 13 |
| Giardia | Direct exam | 13 | 13 |
| Helminths | Direct exam | 13 | 13 |
| Other protozoa | Direct exam | 13 | 13 |
| <i>Spironucleus</i> sp. | Direct exam | 13 | 13 |