Induction and Monitoring Of Experimental Autoimmune Encephalomyelitis (EAE) In Rodents

IACUC Standard Procedure
Effective Date: August 2023

UCSF Office of Research
Institutional Animal Care and Use Program

Purpose: There are numerous scoring systems for assessing and treating rodent models of experimental autoimmune encephalomyelitis (EAE) and related demyelinating diseases. The purpose of this guideline is to provide a commonly used system which both assures animal welfare and fulfills investigators' scientific objectives.

Note: If lab-specific scoring systems are used, the lab must clearly define clinical signs and management in the IACUC protocol and post this information in the housing room. All EAE animals must be listed in the protocol as Category E.

Description of procedure: EAE is a demyelinating disease of the central nervous system (CNS) that serves as a model of CNS autoimmune disease, most often human multiple sclerosis (MS). EAE can be induced by genetic manipulation or by administering combinations of CNS components (cells and/or peptides) and Complete Freund's Adjuvant (CFA), often with pertussis toxin or viral/pathogen inoculation, or by administration of myelin-specific T cells in the absence of CFA. These combinations result in a complex spectrum of acute, chronic and relapsing-remitting disease courses that most often result in varying degrees of progressive ascending paralysis.

Recordkeeping: Due to the extreme variability of in the onset and progression of clinical signs and disease course, close monitoring and provision of supportive care are necessary for EAE animals. All daily monitoring, assessments, scoring (EAE and BCS), weights, experimental inoculation(s), supportive care and treatments must be recorded in a monitoring logbook/record maintained by the laboratory with the most current information and kept in the housing room, either as paper records or a QR code link to electronic records. Notations in the record must also include name of observer(s), date and time of log entry. An example EAE logbook sheet is available as an editable document on the IACUC website. If the protocol is IACUC approved for Researcher Care, refer to IACUC Policy 'Laboratory Housing and Care of Animals by Researchers Researcher Care' for additional detail regarding daily monitoring, documentation requirements, request forms, and cage stickers.

Veterinary Assessment: As disease progresses, veterinary assessment of the animal condition is essential. LARC veterinary staff may also evaluate animals daily. This will include gentle handling when needed to fully assess the animal. If the LARC veterinarian mandates euthanasia for humane reasons, the animal must be euthanized.

EAE Score	Clinical Signs	Required Recordkeeping and Communication	EXPERIMENTAL MONITORING FREQUENCY Signs to monitor	Intervention
0	Clinically normal	• Each animal must be individually identified. • Label each cage: "EAE" and date of EAE induction • If Researcher Care[4] is IACUC approved, label cages 'Lab Care'[4]	**DAY 0** = EAE induced = immune potentiator injected • Record baseline bodyweight[1] **AT LEAST 3X/WEEK** • EAE score. Initiate prior to expected onset of clinical signs to monitor for disease progression. • **NOTE**: if protocol is IACUC approved for Researcher Care[4] of EAE animals, lab is still required to check animals daily.	**Optional** • Additional water source (e.g. long sipper tubes, gel product, etc.) • Moistened food pellets on cage floor or high calorie nutrition
1	Weak tail		**AT LEAST 3X/WEEK** • EAE score • Signs in Table 1.EAE Adverse Effects **OPTIONAL** • Bodyweight[1] or Body Condition Score (BCS), 3x/week	**Optional** • Additional water source (e.g. long sipper tubes, gel product, etc.) • Moistened food pellets on cage floor or high calorie nutrition
2	Weak hind limbs		**DAILY**[2] • EAE Score • Signs in Table 1. EAE Adverse Effects **OPTIONAL** • Bodyweight[1] or BCS, daily	**Required** • Additional water source (e.g. long sipper tubes, gel product, etc.) • Moistened food pellets on cage floor or high calorie nutrition
3	One hindlimb paralyzed		**DAILY**[2] • EAE score • BCS • Dehydration: evaluate visually or check skin turgor (see Table 1. EAE Adverse Effects). • Urinary incontinence lesions: dermatitis, urine scald, penile prolapse • Additional signs in Table 1. EAE Adverse Effects, **OPTIONAL** • Bodyweight[1] daily	**Required** • Cage set-up (see photo last page) • Long sipper tube, gel products • Food on floor or high calorie nutrition • If dehydrated: at least 1 mL (or volume per LARC vet) of pharmaceutical grade sterile 0.9% Saline or Lactated Ringer's, SC or IP, once per day. Give more frequently as specified per LARC veterinary requirements. • Urinary incontinence lesions: Contact LARC vet staff.[3] LARC vet staff will assess severity on a case-by-case basis. Treat, monitor, and euthanize at the discretion of LARC veterinary authority. • Euthanize: if urine scald or infection are non-responsive to treatment, and/or penile prolapse. **Recommended** • House with healthy animals for thermoregulation. • If animal has difficulty ambulating, you may give extra nesting material (enviro-dry handful or 2-3 nestlets). C*aution*: animals can get tangled in it.

EAE Score	Clinical Signs	Required Recordkeeping and Communication	EXPERIMENTAL MONITORING FREQUENCY Signs to monitor	Intervention
3.5	Both hindlimbs paralyzed		**DAILY**[2] • EAE score • BCS • Signs in Table 1. EAE Adverse Effects, **OPTIONAL** • Bodyweight[1] daily	**Required** • Hydration gelpack on cage floor • Moistened food pellets on cage floor or high calorie nutrition • At least 1 mL (or volume per LARC vet) of warmed pharmaceutical grade sterile 0.9% Saline or Lactated Ringer's, SC or IP, once per day. Give more frequently as specified per LARC veterinary requirements. • Urinary incontinence lesions: Contact LARC vet staff[3]. LARC vet staff will assess severity on a case-by-case basis. Treat, monitor, and euthanize at the discretion of LARC veterinary authority. • Euthanize: if urine scald or infection are non-responsive to treatment, and/or penile prolapse.
4	Both hindlimbs paralyzed with weak front limbs	Notify LARC vet staff[3] of score same day	**DAILY**[2] • EAE score • BCS • Signs in Table 1. EAE Adverse Effects **OPTIONAL** Bodyweight[1] daily	**Required** • **CONTINUE** all care described above.
4.5	Both hindlimbs plus one front limb paralyzed, or non-functional movement (can move but can't get to food/water)	• Document date and time EAE score 4.5 first seen. • Immediately notify LARC vet staff[3] of score.	**TWICE DAILY**[2] • EAE score • BCS • Signs in Table 1. EAE Adverse Effects **OPTIONAL** • Bodyweight[1] twice daily	**Required** • Continue all care described above but **increase fluid therapy to twice per day**. • Any animal that does not remit to Score 4 or lower within 24 hours must be immediately euthanized.
5	Both hindlimbs and both front limbs paralyzed, and/or moribund			**Required** • Euthanize as soon as possible within 1 hour. • LARC may euthanize if moribund or lab has not done it within 1 hour.

[1] Recommended baseline weight calculation is average of at least 3 weights measured over a one-week period (5-7 consecutive days). Euthanasia is required if animal has 20% acute weight loss from baseline weight over 24 hours, or a cumulative 25% maximum weight loss.

[2] Daily monitoring, scoring, treatment and documentation of animals applies to weekends and holidays and is the responsibility of the Principal Investigator. Records must be kept in the housing room, either as paper records or a QR code link to electronic records and provided to IACUC or LARC staff upon request.

[3] Email LARC vet staff at healthcheck@ucsf.edu or verbally notify LARC staff in room.

[4] Researcher Care (aka 'Lab Care') must be described and approved in IACUC protocol prior to implementing. LARC will open cages to examine animals at any point based on veterinary assessment. LARC vet may jointly assess with lab (to reduce animal stress) as schedule allows. If the protocol is IACUC approved for Researcher Care, refer to IACUC Policy 'Laboratory Housing and Care of Animals by Researchers Researcher Care' for additional detail regarding daily monitoring, documentation requirements, request forms, and cage stickers.

Cage Set-up

Required[1]

- Long sipper tube, gel products, and fluid replacement (SQ fluids)
- Food on floor or high calorie nutrition

Recommended

- House with healthy animals for thermoregulation benefits.
- If unable to ambulate well, extra nesting material *may* be helpful (e.g., a handful of enviro-dry or 2-3 nestlets), *however, use with caution*. In some cases, extra nesting material can make ambulation more difficult, i.e. animals can get tangled in nestlets.

[1]Requirements vary per health assessment and scientific goals.