



Hooke Laboratories

Immunization of Mice for Generation of Encephalitogenic T Cells

Recommended protocol for use with:

- Hooke Kit™ MOG₃₅₋₅₅/CFA Emulsion (cat. no. EK-0111)
- Hooke Kit™ [Ser¹⁴⁰]-PLP₁₃₉₋₁₅₁/CFA Emulsion (cat. no. EK-0120)
- Hooke Kit™ MBP₁₋₁₇/CFA Emulsion (cat. no. EK-0130)
- Hooke Kit™ PLP₁₃₉₋₁₅₁ (native)/CFA Emulsion (cat. no. EK-0230)

Summary

Mice are immunized with peptide antigen emulsified in Complete Freund's Adjuvant (CFA) to generate encephalitogenic cells for adoptive transfer EAE.

Mice immunized with MOG₃₅₋₅₅ or MBP₁₋₁₇ antigens according to this protocol (not administered pertussis toxin) will usually not develop EAE. If EAE does develop, it will be milder than when pertussis toxin is used and the onset will be significantly later than in mice injected pertussis toxin.

Materials needed

Qty	Description
1	Hooke Kit™ MOG ₃₅₋₅₅ /CFA Emulsion (EK-0111), Hooke Kit™ [Ser ¹⁴⁰]-PLP ₁₃₉₋₁₅₁ /CFA Emulsion (EK-0120), or, Hooke Kit™ MBP ₁₋₁₇ /CFA Emulsion (EK-0130) Hooke Kit™ PLP ₁₃₉₋₁₅₁ (native)/CFA Emulsion (EK-0230)
10	MOG ₃₅₋₅₅ antigen (EK-0111) – female C57BL/6 mice [Ser ¹⁴⁰]-PLP ₁₃₉₋₁₅₁ antigen (EK-0120) – female SJL mice PLP ₁₃₉₋₁₅₁ (native) antigen (EK-0230) – female SJL mice MBP ₁₋₁₇ antigen (EK-0130) – female B10.PL, PL/J, or anti-MBP TCR transgenic mice

Method

Mice should be 8-16 weeks old, acclimated at your facility for at least 7 days before immunization.

Inject each mouse with a total of 0.2 mL of emulsion subcutaneously, at 2 sites on the back – one on the upper back, one on the lower back - administering 0.1 mL per site.

Note: For SJL mice, inject 0.2 mL at 4 sites – one over each shoulder and one over each hip – administering 0.05 mL per site.

Keep the needle inserted into the subcutaneous space for 10 to 15 seconds after each injection to avoid leakage of the emulsion. Alternatively, a light pull on the syringe plunger will prevent leakage.

Encephalitogenic T cells can be generated by culturing splenocytes from the immunized mice 10 to 11 days after immunization (10 days for SJL mice). See Hooke's Adoptive Transfer EAE protocols for details (<http://hookelabs.com/protocols>).

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