

Mouse CIA – A Visual Guide

For Research Use Only - Not Human or Therapeutic Use

MICE: CHOOSE 1 STRAIN

DBA/1 (DBA) mice **OR** C57BL/6 (B6) mice

- 7-8 weeks old
- SPF housing conditions
- High fat diet



RECOMMENDED ADJUVANTS & COLLAGENS: CHOOSE 1 ADJUVANT + 1 COLLAGEN

Catalog #	Adjuvants
7002	Incomplete Freund's Adjuvant (IFA) (DBA - Booster only)
7008	Complete Freund's Adjuvant (CFA), 1 mg/ml (DBA)
7001	Complete Freund's Adjuvant (CFA), 4 mg/ml (DBA)
7023	Complete Freund's Adjuvant (CFA), 5 mg/ml (B6)

Catalog #	Adjuvants
20011	Chick Type II Collagen, Lyophilized (DBA or B6)
20012	Chick Type II Collagen, 2 mg/ml Solution (DBA or B6)
20021	Bovine Type II Collagen, Lyophilized (DBA)
20022	Bovine Type II Collagen, 2 mg/ml Solution (DBA)

IMMUNIZATION SCHEDULE: CHOOSE 1 OPTION

OPTION 1: Single Immunization Protocol (DBA mice)

Day 0: Subcutaneous injection: Collagen emulsion with CFA: 4 mg/ml *M. tuberculosis*

Day 0 - 28: Score arthritis every 7 days

Day 28 - 56: Score arthritis every 2-3 days: Arthritis incidence 90 - 100%

Day 56: End of experiment.

OR

OPTION 2: Booster Immunization Protocol

Day 0: Subcutaneous injection: Collagen emulsion with CFA: 1 mg/ml (DBA) or 5 mg/ml *M. tuberculosis* (B6)

Day 0 - 28: Score arthritis every 7 days

Day 21: Subcutaneous injection (booster): Collagen emulsion with IFA (DBA) or CFA: 5 mg/ml *M. tuberculosis* (B6)

Day 28 - 56: Score arthritis every 2-3 days: Arthritis incidence 90 - 100% (DBA), 50-70% (B6)

Day 56: End of experiment.

© 2019 Chondrex, Inc. All Rights Reserved, 1.0

PREPARE EMULSION WITH A HOMOGENIZER: DO NOT USE THE DOUBLE-SYRINGE METHOD!

Figure 1.

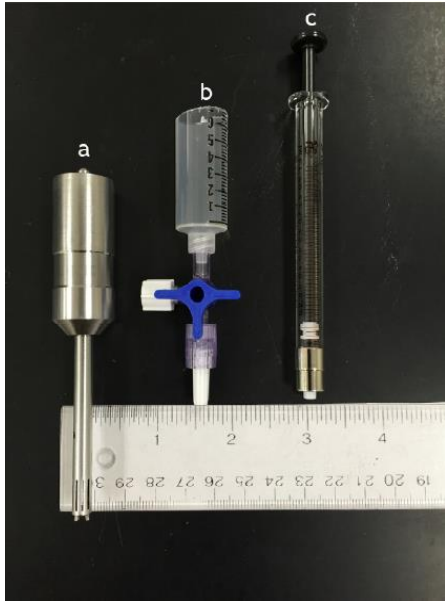
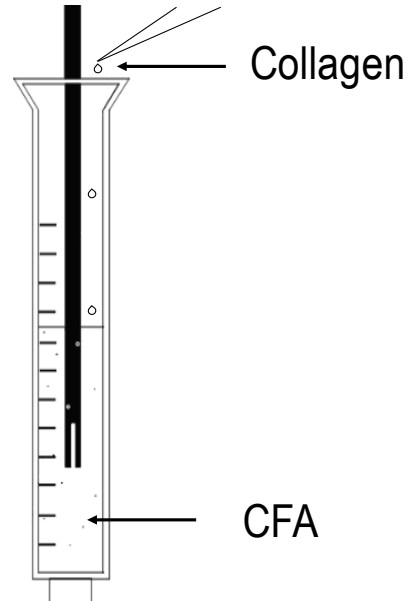


Figure 2.



Figure 3.

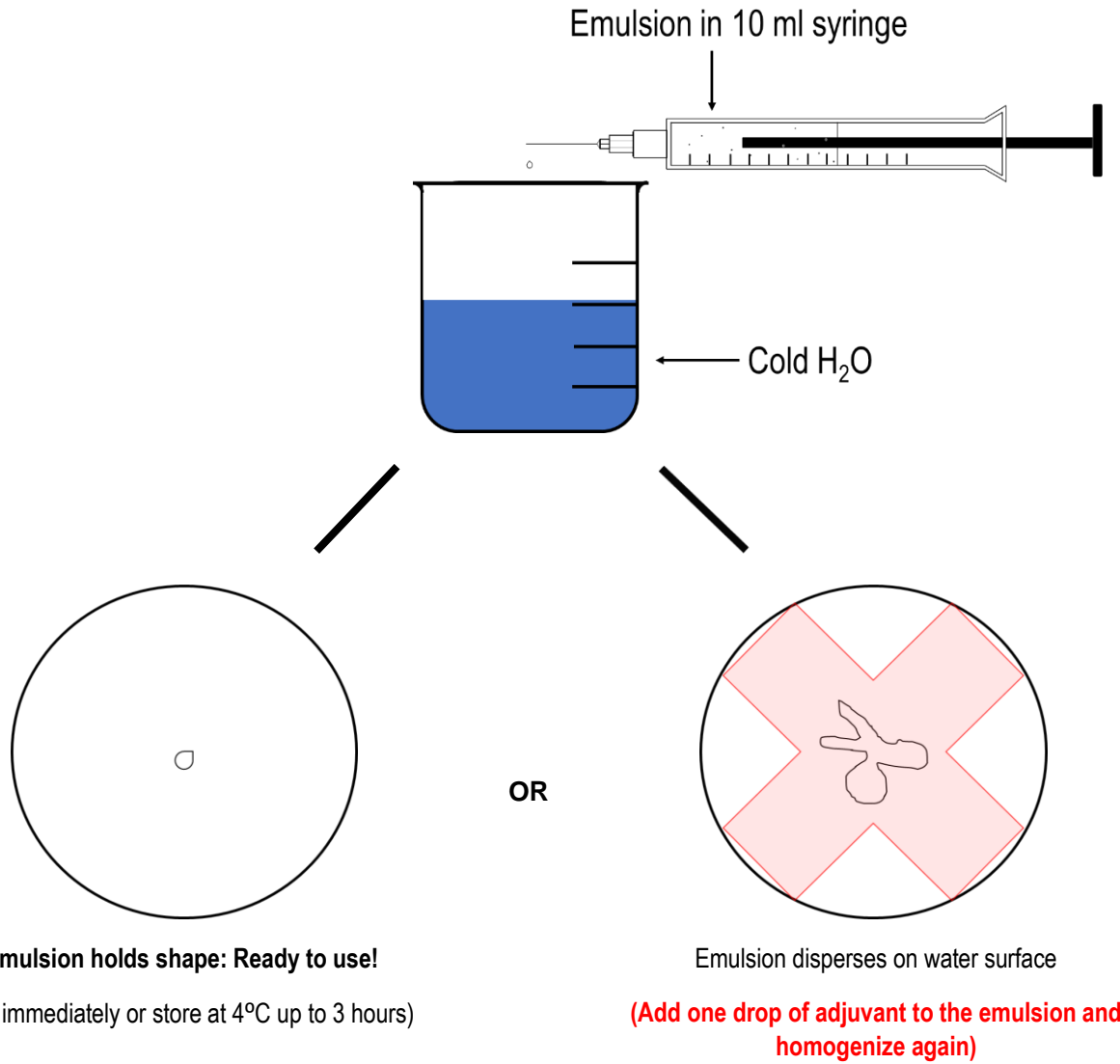


Procedure*:

1. Gather materials (Figure 1. Homogenizer blade (a), 10 ml syringe with a 3-way stopcock (b), and 1 ml Hamilton syringe (c)).
2. Lock stopcock.
3. Keep setup on ice (Figure 2).
4. Add CFA to syringe.
5. Add an equal volume of collagen solution, drop-wise (Figure 3).
6. Mix at 10,000 - 30,000 rpm for 2 minutes. Move the syringe up and down while mixing.
7. Cool syringe on ice for 5 minutes.
8. Mix at 10,000 - 30,000 rpm for 2 minutes. Move the syringe up and down while mixing.
9. Evaluate emulsion with the drop test (next page).
10. Transfer the emulsion to a Hamilton syringe using the 3-way stopcock.

*For more information, please see Chondrex, Inc's [Mouse CIA Protocol](#)

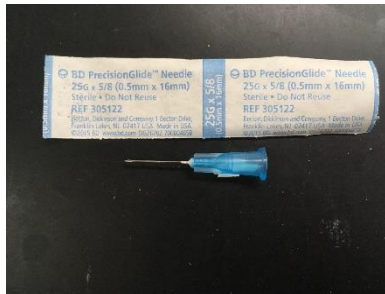
EMULSION DROP TEST:



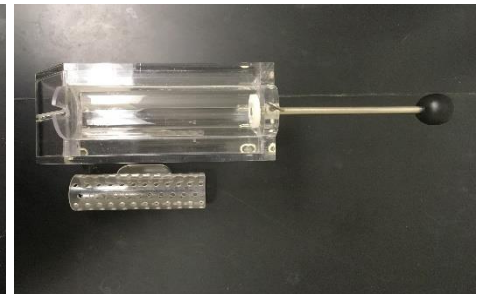
IMMUNIZATION:



(a) Emulsion in Hamilton Syringe
(no air bubbles!)

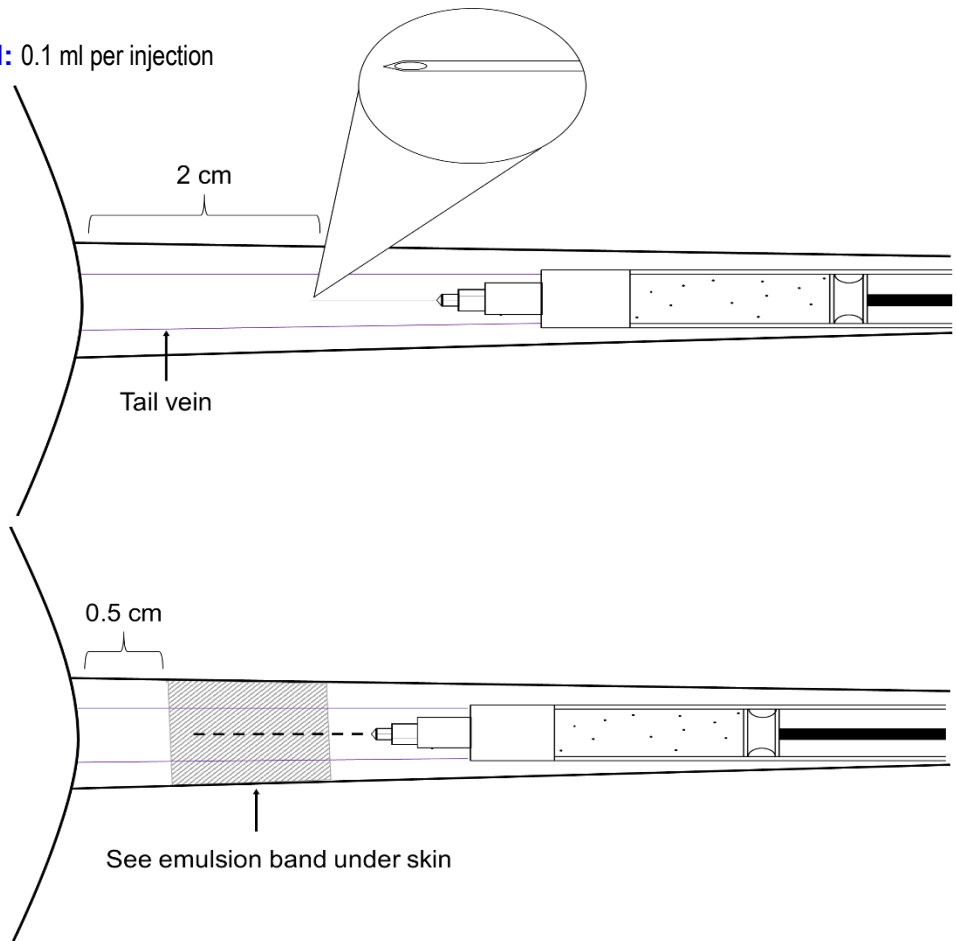


(b) Needle (25 or 27-gauge x 5/8")



(c) Restraining Device

SUBCUTANEOUS INJECTION: 0.1 ml per injection



**DO NOT USE
IP (a) OR IV (b)
INJECTIONS TO
IMMUNIZE MICE!**

