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## Standard Operating Procedure Injections and Immunizations

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### **Intrahepatic injection of cells for mouse reconstitution**

1. Adjust cells to  $10^5$ - $3 \times 10^5$  CD34<sup>+</sup> hematopoietic progenitor cells in 20 $\mu$ l per mouse pup/injection with cold PBS
2. Prepare Hamilton syringes for injection
3. Fix mouse pup by grabbing in the neck
4. Inject animal with 20 $\mu$ l cell suspension intrahepatically on the right side of the abdominal cavity (left side when mouse abdomen is facing you)

### **Intraperitoneal injections of mice**

1. Adjust cells or substances to be injected to 200 $\mu$ l per mouse/injection (up to 10ml/kg body weight per injection)
2. Prepare insulin syringes for injection
3. Fix mouse by grabbing in the neck and pulling back the fur
4. Inject animal with up to 200 $\mu$ l (10ml/kg body weight per injection) cell suspension on either side of the abdominal cavity
5. Mice can be daily injected for up to 4 to 6 weeks

### **Injections via intravenous route**

1. Adjust volume of cells or substances to up to 200 $\mu$ l PBS per mouse
2. Prepare 1ml syringe with 27G needles for injection. Avoid bubbles!!
3. Warm the mouse and place it in a restraining tube.
4. Wipe tail with alcohol pad
5. Inject 200 $\mu$ l of cells or substances carefully i.v.
6. Stop bleeding with gauze pad

### **Subcutaneous injections**

1. Anesthetize mice using the isofluoran-evaporation chamber according to the SOP "Anesthesia"
2. Remove mice from the chamber



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3. Lift the neck skin if the mice with the index finger and thumb of the left hand and carefully inject up to 10ml/kg body weight of liquid into the lifted neck skin or into lateral flanks using an insulin syringe

### **Immunization of mice with tetanus-toxoid or DEC-205 fusion antibody**

1. Prepare endotoxin-free, azide-free fusion antibody in PBS on ice. For TT, mice are injected with 1/50 of a human dose (=10 $\mu$ l). For fusion antibodies 5 $\mu$ g are used per injection.
2. Adjust volume of TT or antibody dose with 50 $\mu$ g Poly(I:C) as adjuvant to 200 $\mu$ l with cold PBS.
3. Prepare 0.5ml insulin syringes for injection.
4. Fix animals by grabbing in the neck and pulling back the fur.
5. Inject animal with 100 $\mu$ l of TT or antibody on either side of the abdominal cavity

### **Immunization of mice with lentiviral particles encoding latent EBV particles via intraperitoneal route**

1. Thaw viral stocks on ice.
2. Adjust volume of planned viral dose to 100 $\mu$ l with cold PBS. 1-5x10<sup>8</sup> recombinant lentiviral particles are used per injection.
3. Prepare 0.5ml insulin syringess for injection.
4. Fix animals by grabbing in the neck and pulling back the fur.
5. Inject animal with 100 $\mu$ l of virus on either side of the abdominal cavity

### **Immunization of mice with lentiviral particles encoding latent EBV particles via intravenous route**

1. Thaw viral stocks on ice.
2. Adjust volume of planned viral dose to 10 $\mu$ l with cold PBS. 1-5x10<sup>8</sup> recombinant lentiviral particles are used per injection.
3. Prepare 1ml syringe with 27G needles for injection. Avoid bubbles!!
4. Warm the mouse and place it in a restraining tube.
5. Wipe tail with alcohol pad
6. Inject 10 $\mu$ l of cells carefully i.v.



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7. Stop bleeding with gauze pad