

Chronic cranial window implantation

1- INJECTION-prepare syringes:

Ketamine/xylazine 4:1 -10 ul/g animal (intra-peritoneal injection –i.p.)

24ml NaCl

4 ml ketamyn

1ml xylazin

Dexamethasone (anti-inflammatory) (4mg/ml in 0.9% NaCl) – 2,5 ul/g animal (i.p.)

Rimadyl (pain killer) – 7ul/gr animal (subcutaneously)

CEFO (cefotaxime, antibiotic) – 2,5 ul/g animal (subcutaneously)

2- PREPARE INSTRUMENT AND PLACE:

Pasteur pipette (sterile)

Cotton swabs (sterile)

Gelfoam – cut it in small pieces and leave in PBS

Sugi

Skalpel (sterile)

Biopsy punches (3-5mm)

Glass for pipettes

PBS

EtOH

Dental cement (glue and cement)

Coverslip (choose the size according to the size of the biopsy punches, from 3 up to 5 mm)

Bepanthene

Drill tip

Metal holder

- Remember: Sterilize medical devices, as well as coverslip.

3- SURGERY PROCEDURE

-Check the mouse is completely anesthetized checking tail, paws and whiskers reflex

-Fix the animal and cover eyes with bepanthene

-Clean the head with 70 % EtOH

-Cut the skin (triangular shape): not too close to the ears not too close to the neck

- Clean the skull with the scalpel and the vacuum cleaner. Remove all the hairs. Use PBS to help you during this procedure.
- Scratch the skull with the scalpel: in this way you increase the area for the final glue and at the end the helmet will be better fixed to the head of the animal.
- Mark window place with the punch (choose the punch according to the size of the window you want, like 3 or 4 mm)
- Drill the window edges till the bone is very thin and transparent
- Add some PBS on the surface to make easier the removal
- Pull gently the skull surface you just drilled (don't pull to the slide): stop the bleeding with gel foam.
- Be careful: Never let the brain get dry; in order to avoid that add a drop of PBS on the surface.
- Wash the cover slip with EtOH and PBS. Place it INSIDE the window
- Fill glass pipette with histological glue, fix coverslip.
- Scratch again the skull, mix cement and make the helmet. To create a good mix of cement and glue use the needle from a syringe and then spread the mixture all over the skull. Once you are close to the window make it more dense to ensure it doesn't spread over the window.
- Glue the holder on the cement mix.

4-AFTER THE SURGERY

- Remove carefully the animal from the surgery setup.
- Place the animal back in his cage.
- Place the cage in the incubator till the animal is awake and able to move.
- Replace the cage and the animal in the animal room.