



Date 20170622

## Standard Operating Procedure Anesthesia

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### **Isofluorane anesthesia**

1. Assemble the induction chamber with the tube for discharging used isofluorane/oxygen mixture and the tube for delivering fresh isofluorane/oxygen mixture.
2. Clean the induction chamber well with 70% EtOH.
3. Place animals in the chamber.
4. Turn on the oxygen supply.
5. Set vaporizer value to 5% for the induction of anesthesia. Reduce to 3% after induction.
6. Set the flow to 2 liters per minute.
7. Wait until respiration rate is 50-60 per minute.
8. Take animal out and turn of isofluorane and oxygen flow.
9. Do not put animals in chambers already filled with gas. Empty chamber and let the gases flow out before starting another anesthesia.

### **Isofluorane anesthesia (IVIS)**

1. Turn on "Evacuation Pump" and the oxygen supply.
2. Turn on gas flow in IVIS Imaging Chamber
3. Turn on the gas flow to the XGI-8
4. Set Vaporizer Value to 0% agent
5. Turn on "IVIS FLOW"
6. Turn on Flow to Induction chamber
7. Set the Vaporizer to appropriate setting for animal induction, typically at 2.5%.
8. Place animal in induction chamber
9. Turn off "chamber on/off" prior to removing animals from induction Chamber
10. Place animals in the nose cones
11. At the end of the procedure: turn the vaporizer to the off position. Turn on the "Chamber on/off" to allow the pure oxygen to flow through the induction Chamber for 5 minutes.
12. Turn off the supply oxygen/the XGI-8/the evacuation pump



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### Injection anesthesia

1. Mice are weighted.
2. Mice are anesthetized by i.p. injection of 0.05 mg/kg Fentanyl, 5 mg/kg Midazolam, 0.5 mg/kg Medetomidin.
3. After onset of narcosis mice are placed on a heating pad to prevent hypothermia.
4. After 5 to 10 min, when surgical tolerance is reached (lack of retraction reflex when pinching the foot between two fingers)
5. After manipulation of mice, animals should be monitored for another 5-10 min for abnormal breathing rate.
6. Abnormal breathing can be counteracted by placing the animal in an upright position and/or immediate antagonisation of anesthesia.
7. Animals are antagonized by i.p. injection of 1.2 mg/kg Naloxon, 0.5 mg/kg Flumazenil, 2.5 mg/kg Atipamezolin. Fentanyl and Midazolam should be antagonized first by Naloxon and Flumazenil. Atipamezol should be administered 10 minutes later in case the mice are not awoken yet.

### Injection anesthesia (alternative)

1. Mice are weighted.
2. Mice are anesthetized by i.p. injection of 100 mg/kg Ketamin, 14 mg/kg Xylazin.
3. After onset of narcosis mice are placed on a heating pad to prevent hypothermia.
4. After 5 to 10 min, when surgical tolerance is reached, animals are allowed to aspirate 25-50µl of influenza A virus.
5. Observe animals for another 5-10 min for complete aspiration of fluid and abnormal breathing rate.
6. Abnormal breathing can be counteracted by placing the animal in an upright position and/or immediate antagonisation of anesthesia.



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7. Animals are antagonized by i.p. injection of 5 mg/kg Atipamezolin. Atipamezolin should not be administered within the first 60 minutes past the initial Ketamin administration as it could source a Ketamin-hangover induced catalepsy.