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Date: 14.07.2020	Isoflurane Anesthesia Isoflurane	LTK-TRT-13-B-EN Version: B

This SOP replaces:	Date: 11.07.2015 Version: A	
Reason for Change:	Commission comments to colleague: 1. Suction may only be started when the animal is at the nosepiece 2. The flow for the induction box must be at least 1 liter. 3. For induction, the isoflurane evaporator must be set to 5%. 4. As soon as the animal is connected to the nose piece, even if it is only for 2 minutes, the animal must be placed on a heating pad.	
Related SOPs:	none	
Indication of Use:	Procedures that require the animal to be unconscious.	
Aim of SOP: Distribution: Attachments:	This procedure describes how to perform anesthesia by isoflurane 1. Server	
Generated at: 14.7.2020	Checked and approved at: 14.7.2020	
by: Thorsten Buch	by: Philippe Bugnon	

Responsible Persons: Any person with Modul 1 and registered on a particular animal permit
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Method: inhalation of isoflurane in conjunction with oxygen

Principle of Method:

1. Inhibits NMDA glutamate receptors, and potassium channels, inhibits Ca ATPase. Binds to GABA receptors and glycin receptors ATPsynthase D and NADH dehydrogenase

Calibration: ensure that the nebulizer is maintained regularly

Machine:


Laminar flow/changing station
Isoflurane setup
Optional: Heating pad

Reagents:

1. Isoflurane
2. Oxygen
3. Optional: Vit A ointment or Humigel for long anesthetics


Safety:

1. If possibly pregnant: do not use isoflurane!!!
2. Follow the rules for the animal house
3. If machine uses coal filter: check weight before and after use. The weight should be marked as soon as an increase of more than 40 g is measured. And as soon as the weight is less than 40 g from the limit, mark the weight as soon as an increase of 10 g is measured. If necessary, change filter.

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Method Description:

1. Check the level of oxygen to be sure that you will have enough oxygen for the duration of your experiment: Calculation of the oxygen available: pressure (bar) x volume of the oxygen bottle (liters) = how many minutes available (with a flow of 1 liter / minute). Example: 100 (bar) x 10 liter bottle = 1'000 minutes of oxygen available.
2. Check the weight of the coal filter: the limit of use is written on the filter. If the weight is over this limit, replace the filter before the anesthesia. A filled filter will not absorb the anesthetic agent which will accumulate in the tube and provokes an overdose and kill your animal.
3. Check if the evacuation pump of the coal filter is working.
4. Check the level of isoflurane and refill if necessary before opening the oxygen supply.
5. Check all connections (coal filter, oxygen to vaporizer, vaporizer to induction box and mask
6. Check the presence of the pin in the mask (necessary for mice)
7. Turn on oxygen supply.
8. Turn on the gas flow to the vaporizer (1000 ml/min).
9. Set vaporizer to 0% agent
10. Turn on flow to induction chamber
11. Set the vaporizer to appropriate setting for induction of anesthesia, typically at 5%.
12. Place animal in induction chamber, wait 2-3 minutes for anesthesia. Check the righting reflex by placing the animal on the back (through tilting of chamber, without opening the chamber).
13. Set vaporizer to 2% and flow to 650 ml/min to maintain anesthesia. If lower than 600 ml/min, the percentage of isoflurane in the flow will be unstable.
14. Turn flow to anaesthesia mask (nose cone).
Place animals on nose cone and turn on immediately the evacuation pump of the coal filter.
15. Check the respiration rate (stable, regular and low) and adapt the percentage of isoflurane if necessary.

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16. Additional important points:

- a. Your animal must be placed on a heating pad set to 37°C if the anesthesia lasts for more than 2 minutes.
- b. If anesthesia lasts for more than 5 min: protect eyes with ointment (Vit A ointment or Humigel)

17. At the end of the anesthesia, turn the vaporizer to the off position to deliver to the animal only pure oxygen, until the animal starts to move.

18. Place your animal in a recovery cage.

19. Turn off the oxygen supply and the evacuation pump.

20. Empty the isoflurane from the induction box.

21. Check the level of oxygen and the coal filter, replace if necessary


Weight the Aldasorber (coal filter), if more than the limit (written on the filter) ! replace it. Write the weight and the date as soon as the difference is more than 40 grams. And within 40 grams from the limit, write any difference of 10 grams with the date.

Controls:

control pinch reflex (skin between toes of the hind limb) before start of surgery

Documentation:

Lab book, Score sheet SOP scoring post-application (depending on permit)

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Problem management:

Case 1: the animal does not sleep

- vaporizer empty?
- O₂ cylinder empty?
- Connections ok?

Case 2: induction OK, but later the animal does not sleep anymore

- nose pin lacking?
- Nose mask defect?
- O₂ flow too low (< 600 ml/min)?

Case 3: Induction OK, then later the animal dies

- Isoflurane still on induction value?
- extraction pump ON ?
- adsorber full?
- warming pad ON?

Report any other adverse event to your supervisor or vet

Literatur: