A Low Cost Face Mask for Inhalation Anaesthesia in Rats

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Summary
Inhalation anaesthesia in small laboratory rodents has become very popular in Biomedical Research. A common method to deliver volatile anesthetic agents is through a face mask. The main disadvantage of this method is the exposure of personnel to anaesthetic agents. The authors describe a low-cost face mask, made with materials commonly accessible in the operating room that can be connected to an anaesthetic machine eliminating personnel exposure to hazardous levels of anaesthetic agents and allowing for effective adjustments to the anaesthetic depth.

Introduction
The use of volatile anaesthetic agents in small laboratory rodents has become very popular in Biomedical Research. The main advantages of inhalation anaesthesia are the decreased induction and recovery times and the increased control over the anaesthetic depth. These features make such an anaesthetic technique the preferable method of anaesthesia for rat surgery *(Dubley et al., 1975; Guthy, 1975).*
Various methods of delivering volatile anaesthetic agents to small rodents have been described in the literature *(Flecknell et al., 2007)*. One of the most commonly used delivery systems is the anaesthetic-delivery chamber. The chambers are constructed with a fresh gas (anaesthetic gas and oxygen) inlet as well as with an outlet for exhaled and excess gases. The animals, after loss of consciousness, are removed from the chamber and anaesthesia is maintained by using a face mask connected to a non-rebreathing circuit. Inevitably, opening of the chamber's door, in order to remove the animal, causes release of anaesthetic gas. Therefore, one of the major disadvantages of using the anaesthetic-delivery chamber is the exposure of personnel to the anaesthetic gas, even when a hood is being used *(Wixson and Smiller, 1997; Flecknell et al., 2007)*. Face masks, apart from maintenance of anaesthesia, are also used for induction of anaesthesia *(Flecknell et al., 2007)*. Lack of scavenging systems make the use of face masks unsafe and the use of special operating tables with built in scavenging systems are highly correlated with patient hypothermia which can have detrimental results *(Li et al., 2001; Smith and Bolon, 2006)*.
Other reported delivery methods of inhalant anaesthetics require endotracheal intubation. This method enjoys wide acceptance but demands skilled personnel and special equipment. Rat intubation is more difficult not only because of the small size of the palate and oropharyngeal cavity, tiny larynx and epiglottis, but also because of the lack of adequate equipment *(Boersma and Wieringa, 1982; Schaefer et al., 1984)*. Several methods of endotracheal intubation of various degrees of complexity have appeared in the literature *(Stark et al., 1981; Thet, 1983; Yasaki and Dyck, 1991; Ordodi, 2005)*, and breathing can be either spontaneous or controlled with the use of positive pressure ventilation. For spontaneous breathing the tracheal tube is connected to an open, non-rebreathing system and a scavenger is used for the collection of the exhaled gases providing personnel safety. For positive pressure ventilation, a closed breathing system is connected to a ventilator. Intubation must follow premedication and induction of anaesthesia with injectable or

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volatile agents *(Stark et al., 1981; Schaefer et al., 1984; Jou et al., 2000)*.

The purpose of this study was to describe a low-cost face (LCF) mask which was used during surgical implantation of a telemetry device in rats, made with materials commonly accessible in the operating room that can be connected to an anaesthetic machine eliminating personnel exposure to hazardous levels of anaesthetic agents and allowing for effective adjustments to the anaesthetic depth.

Materials and Methods

The study was performed in the animal facility of the Center for Experimental Surgery of the Foundation for Biomedical Research of the Academy of Athens. The facility is registered as a "breeding" and "experimental" facility according to the Greek Presidential Decree 160/91, which harmonizes national legislation with the European Community Directive 86/609/EEC on the Protection of Animals Used for Experimental and Other Scientific Purposes *(Council of European Communities, 1986)*.

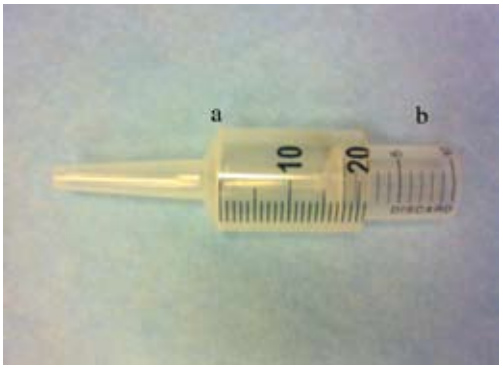

Figure 1. Syringe B is inserted into syringe A.
a: A 50 ml syringe, b: A 20 ml syringe.

Face mask description

A 50 (Becton Dickinson Plastipak, Luer Slip, Ireland) and a 20 ml (Medi-Hut International, Korea) syringe were cut at the 22 (syringe A) and 12 ml (syringe B) mark respectively and syringe B was inserted into syringe A (**Fig. 1**). A 3 mm hole was cut at the surface of the suction tube (CH 24, 210 cm, Pharmaplast, Unomedical A/S DK,) with the use of a No11 surgical blade and a 3 mm oxygen connecting tube (Pennine Healthcare OC-7000, UK) was inserted in the suction tube through the created hole (**Fig. 2**). The syringes were connected to the proximal end of both tubes as illustrated in F**igure 3**. An appropriately cut rubber gasket of a syringe plug was wedged between the outer surface of syringe B and the inner surface of syringe A (**Fig. 4)**. Finally the distal end of the oxygen tube was connected to the vaporizer outlet and that of the suction tube to the incorporated scavenging system **(Fig. 5**). All the

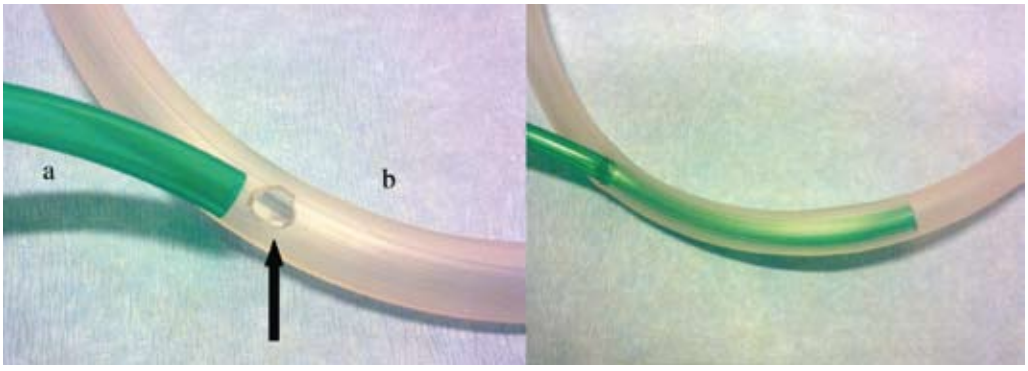

Figure 2. A 3 mm hole (arrow) cut at the surface of the suction tube, through which the oxygen delivery tube is inserted.
a: Oxygen delivery tube, b: suction tube.

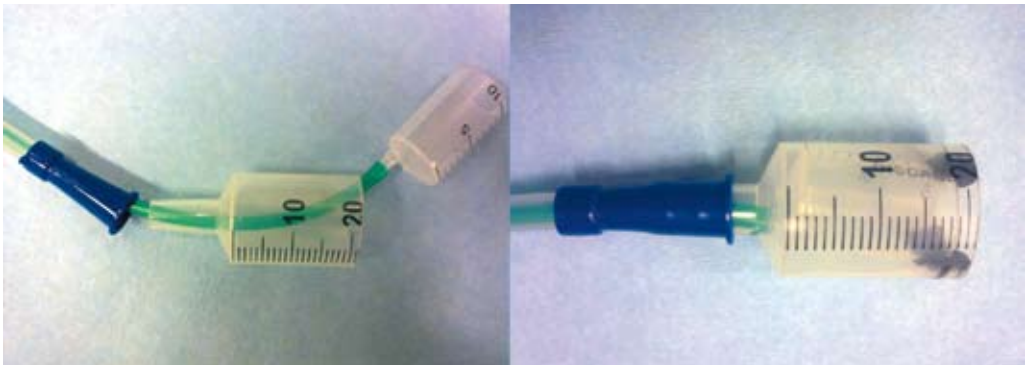

Figure 3. By pulling the oxygen delivery tube all different parts of the face mask get into place.

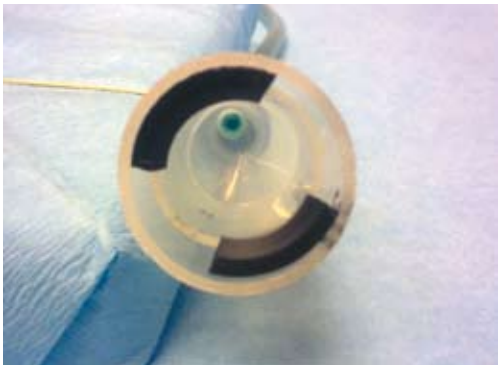

Figure 4. Rubber gasket.

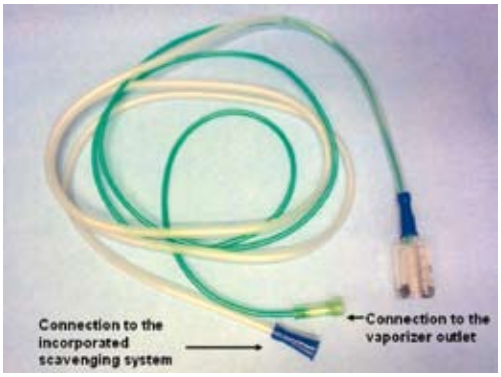

Figure 5. The LCF mask with the breathing system.

materials that were used for the LCF mask are listed in Table 1.

Table 1. Materials needed for the construction of the face mask.

- 1 Connecting tube with 2 funnels for suction
- 1 Oxygen delivery tube 1.8 m F/F 3 mm cross section
- 1 Catheter tip syringe of 50 ml
- 1 Plastic syringe of 20 ml
- 1 rubber gasket of a syringe plug of 50 ml
- 1 flowmeter

Animals

The animals were maintained according to the Guide for the Care and Use of Laboratory Animals and the relevant recommendations of the European Commission on the care and use of laboratory animals *(Council of European Communities, 1986; European Commission, 2007)*.

The animals were pair housed in H-Temp™ polysulfone type III individually ventilated cages (Sealsafe™, Tecniplast, Milan, Italy), with 75 air changes per hour (ACH) under positive pressure with *ad libitum* access to tap water in drinking bottles and an irradiated vacuum-packed pelleted chow that contained 18.8 % protein, 6.0 % fat, 3.8 % fiber,

5.9 % ash (Teklad 2918, Harlan, Italy). All cages were kept in animal rooms under a 12:12h light/dark cycle with lights on at 0700, a room temperature of 24 ± 2°C, a relative humidity of 55 ± 10%, a light intensity of 300 Lux that was measured one meter above the floor in the middle of the room, and a positive air pressure of 0.6 Pa. All animals were regularly screened using a health-monitoring program, in accordance to Federation of European Laboratory Animal Science Associations' recommendations *(Nicklas et al., 2002)*.

The mask was used to maintain anaesthesia in 21 Wistar rats weighting from 300 to 350 g, during surgical implantation of a telemetry device for heart rate monitoring (Data Sciences International, MN, USA). The procedure was reviewed and authorized by the veterinary service of the Prefecture of Athens, as required by the Greek legal requirements for animal experimentation to the European Union Directive 86/609 of the Council *(European Commission, 2007)*. No animal was used in order to evaluate the proper function of the LCF mask in particular.

In all cases anaesthesia was maintained with isoflurane (Forenium, Abbot, Italy) in oxygen delivered via the LCF mask, and a fresh gas flow of 1 L/min. Isoflurane was administered using an animal anaesthesia system (MDS Matrx, England). Initially the vaporizer was set at 4% until the surgical plane of anaesthesia was achieved, and then the setting was reduced at 2.5-3%. Heart rate and oxygen saturation were measured during anaesthesia as well as during the recovery period (Mouse Ox, Starr™, Life Science Corp., PA, USA). Induction, anaesthesia and recovery time were also recorded.

Results

In all 21 cases the induction time to anaesthesia was 1.8±0.2 (SD) minutes, the anaesthesia time was 14.6±1.6 minutes and the recovery period was approximately 1.7±0.6 minutes. During anaesthesia, heart rate variability was 324.4±26.5 beats/min and the mean oxygen saturation was recorded at 95%. No animal died during the procedure. None of the participants (two surgeons and an animal technician) could smell any isoflurane during the surgical procedure.

Discussion

One of the major disadvantages of anaesthesia using a face mask is pollution of the surgical theatre *(Smith and Bolon, 2006)*, since effective gas scavenging can be difficult. In order to overcome this limitation several authors have described different face masks that can be used for induction of anaesthesia in rats *(Dubley et al., 1975; Levy et al., 1980)*. Compared with the face masks already described, the LCF mask is of low cost, constructed by materials commonly available in the operating room. Based on our results, the use of the LCF mask for the surgical implantation of a telemetry device was effective and safe, since the induction time was normal and all animals had an acceptable plane of anaesthesia during the procedure, which suggests that the LCF mask can be safely used to provide anaesthesia to rats.

Although the possible isoflurane emissions were not detected by analyzing the atmospheric air in the surgical room, based on Smith and Bolon (2005) concentrations of 1.7 ppm of isoflurane can be smelled by people. The ability to connect the LCF mask with an active disposal scavenging system or an absorber keeps the operator's breathing zone free from hazardous levels of isoflurane gas, taking into consideration that in our study none of the personnel could smell any isoflurane. Although it is not yet established a recommended exposure limit for isoflurane, since it was not included in the criteria of National Institute of Occupational Safety and Health[1], 2 ppm is applied to newer halogenated agents *(Hoerauf et al., 1996)*. Using objective criteria and based on the results of Smith and Bolon (2005), we assume that

[1] National Institute for Occupational Safety and Health (NIOSH). *In Criteria for a Recommented Standard : Occupational Exposure to Waste Anethetic Gases and Vapors.* Washington, DC: Public Health Service (US Department od Health, Education and Welfare, Centres for Disease Control), US Government Prinitng Office Occupational Safety and Health Administration.

the concentration of isoflurane in the surgical room was less than 2 ppm during our study.

In conclusion, induction of anaesthesia in rats with the use of the LCF mask is a relatively simple method and based on our practical experience can be easily used, mainly for low to medium severity surgical procedures. The system proved reliable and cost less than other commercial systems, while personnel safety was assured since the system can also be connected to the central vacuum circuit to keep the operator's breathing zone free from hazardous levels of isoflurane gas.

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