genic laboratory mice are expensive, and loosing animals during the first steps of inves-
tigation because of a poor airway control may represent a significant additional cost. Many
methonds of tracheal intubation have been proposed for small rodents, and some will be
discussed later in this paper.

We present a video-assisted endoscopic
method of tracheal intubation in mice using a
small bore, 0° arthroscope connected to a
monitor for the visualization of the glottis, a
dullethylene (PE) tube as a cannula for intu-
bation and a guide wire to facilitate its intro-
duction across the vocal cords.

We are using this technique with more than
satisfactory results in a still in progress study on
new generation volatile anaesthetic agents. The
study design includes repeated accurate mea-
surements of several laboratory values over an
eight-week period. This technique is rapid,
safe, and there is no need of a specifically
skilled operator, but only a good knowledge of
mice upper airway anatomy. Mice, on the con-
trary of more invasive airway control tech-
niques, can be re-intubated without any major
complication.

Materials and Methods

Up to may 2004, we used 36 male animals,
aged 8-10 weeks, weight 25-28 g, belonging to
laboratory mice strains CD-1 (50%) and
C57BL/6 (50%) (Charles River, Calco, Italy).
A ll mice were maintained under conventional
housing and feeding conditions in accordance
with EC directives. Study design conforms to the Guide for the Care and Use of Laboratory Animals (NIH 85-23, revised 1996). Atmosphere was obtained administering intraperitoneal a mixture of ketamine (100 mg/kg) and xylazine (5 mg/kg). Absence of both swallowing and righting reflexes has been assumed as a criterion of deep anaes-

**Figure 1.** The Olympus mod. A 7002 straight, small bore (OD = 1.7 mm, length = 58 mm) arthroscope with its connection to the light source.

**Figure 2.** The arthroscope is inserted into the mouth of an anaesthetized mouse in order to allow larynx aditus visualization.

**Figure 3.** After visualization of the aditus, the 1.1 mm OD PE cannula is inserted over the guide wire as an endotracheal tube into the trachea.
A new method of orotracheal intubation in mice

A warmed normal saline pad was placed under supine anaesthetized mice, with head and neck extended, in order to maintain a constant physiologic body temperature.

A straight, small bore arthroscope [outer diameter (OD) = 1.7 mm, 0°, length = 58 mm] (Figure 1), connected to a light source and a videocamera (all instruments and devices have been supplied by Olympus Italian branch), was inserted to the left of incisors down to the base of the tongue to allow visualization of the larynx aditus and rhythmic opening of the vocal cords during spontaneous respiration (Figure 2). Secretions, if present, were removed by a 1.1 mm OD PE cannula, connected to a syringe.

A soft-tip, stiff 0.4 mm OD guide-wire (Arrow Int, PA, USA) was inserted 3-4 mm across the vocal cords during inspiration in order to facilitate the introduction of a 30 mm long, 0.70 OD PE cannula over the guide (Figure 3). Figures 4a-b show the larynx aditus as it appears on the monitor before (Figure 4a) and after (Figure 4b) the insertion of the guide wire. After removing the guide-wire, the animal was connected (Figure 5) to a specifically designed pulmonary ventilator (Rodent Ventilator mod. 683, Harvard, USA) that was set, according to Brown et al., at a tidal volume of 200 µl and at a respiratory rate of 110 breaths/min with a O₂/air 50% mixture and a flow rate of 1 l/min.

Figure 4. A-B. These images, taken with the aid of a videocamera, show the larynx aditus before and after the insertion of the guide wire. Note that the latter is a specifically designed guide for a very thin 24 G arterial cannula.

Figure 5. The mouse as it finally appears connected to the pulmonary ventilator.
After completing the scheduled steps of the study, and after animals involved in have been sacrificed with an intraperitoneal overdose of Tanax, a post-mortem examination has been carried out on 10 of them, in order to detect any possible injury and/or lesion of the upper airway directly connected to the intubating procedure.

**Results**

Despite the anterior location of the glottis, the straight fibre-optic arthroscope allowed good intubating conditions; the success rate, evaluated by the direct visualization and video recording of the introduction of the tip across the vocal cords, was 100% with a mean time of 3 ± 0.4 min. No additional difficulty was observed when mice were re-intubated during the following 8 weeks of the study, and the success rate was 100% again, with similar intubating times.

No major complications resulted. Moderate mucosal bleeding (enough to dirty but not to darken the lens of the arthroscope) has delayed the procedure in three cases, but did not compromise the success of intubation.

Post-mortem examination showed only superficial abrasions without mucosal oedema.

**Discussion**

Tracheal intubation in laboratory animals is indicated in studies where controlled pulmonary ventilation is necessary and in longitudinal studies requiring recovery from repeated general anaesthesia. Moreover, tracheal intubation allows several procedures, such as the instillation of a variety of substances in the lung, pulmonary function measurements and other evaluations, e.g. the use of radiological imaging.

On most medium-sized animals, such as rabbits and dogs, tracheal intubation can quite easily be performed with infant-sized laryngoscope and endotracheal tubes; in smaller animals, such as mice, hamsters, rats and guinea pigs, the technique is more difficult, both because of their size and their anatomic peculiarities.

Ethical concerns of several researchers and animal rights advocates have caused a strong reduction of studies involving primate species and even dogs and cats. Therefore, it is not surprising that the appearance of smaller size laboratory animals in scientific experiments is increasing.

As stated by Jou et al, it seems reasonable to focus some research on techniques and devices introducing simplification and speed in modern animal studies.

Many methods of tracheal intubation in smaller size mammalians have already been described in the literature, and a variety of devices, often self-made, have been proposed; for a selected list of references see Jou et al again.

Surgical methods, both tracheostomy and exposure of the larynx by dissection, are invasive, the postoperative recovery may be too long for the design of the study, and scares and retractions, becoming from surgical approach, do not usually allow repeated procedures. Furthermore, driving the tracheal cannula into the aditus in a blind way, as described when larynx is surgically exposed, increases the risk of taking a false path.

In our study, we do not perform invasive procedures, all materials are commercially available and there is no need of specific training, but a good knowledge of animal anatomic peculiarities is enough for the purpose.

The method of video-assisted intubation of mice we have herein described is rapid, reliable and safe; above all, it is suitable for repeated tracheal intubations.

The cost may be high, but we do not think it is prohibitive.

**References**


