

## Endo Solutions

### Implant preparation

Rats were subcutaneously implanted with silastic implants at the nape of the neck. These implants were either empty or filled with steroid hormones and were of a length aimed at maintaining normal circulating levels, as reported previously (Williams and Lipner, 1982; Albert et al., 1991; Suzuki et al., 1995; Lerant and Freeman, 1998).

Estrogen implants (10% in Sesame oil) were 30 mm in length,

Estrogen (10%) 17-estradiol dissolved in Sesame oil).

Progesterone implants (10% in Sesame oil) were 30 mm in length.

The implants were prepared the day before surgery with silastic tubing (Dow Corning, i.d. 1.98 mm, o.d. 3.18 mm; Futuremedics Australia, Balwyn, VIC, Australia) that was cut to the required length plus an additional 6 mm. Each end of the tube was sealed with 3 mm of silicon sealant.

Before the implant was inserted, it was immersed in 100% ethanol and then soaked in sterile saline (0.9% sodium chloride; Baxter Healthcare) for 10 min.

# Endo Solutions Cannulation

## Anesth

**Ketamine/Xylazine** (0.1 ml (of 87 mg ketamine and 13 mg xylazine per ml) per 100 g body weight.

Initially give 0.1 ml IP and a second 0,1 ml IM after the animal is down.

Anesthesia&analgesiaGDL2.doc

*RAT*

ANESTHETIC	ROUTE	DOSE (Induction)	Duration of Anesthesia (min)	Comments	REF
Isoflorane	Inhalation	3-5%			
Halothane	Inhalation	3-5%			
Ether	Inhalation	To effect			
Pentobarbital	IP	30-50 mg/kg	15-60	light anesthesia	1, 2
Ketamine	IP, IM	50-100 mg/kg		deep sedation/immobilization	1, 2
Xylazine	IP	1-5 mg/kg		light-heavy sedation, analgesia	2
Ketamine + Xylazine	IP	40-100 mg/kg + 5-10 mg/kg	20-30	surgical anesthesia	1, 2

**Alpha-Chlorolose** (56 mg/kg body weight) and **Urethane** (56 mg/kg body weight) will be infused via the catheter for long term anesthesia.

0.6 g Chlorolose in 20 ml saline

6 g Urethane in 20 ml saline

Initial dose is 0.16 ml/ 100 g Bdy Wt in canulae or 0.5 ml/300 gm Bdy Wt

Maintenance dose 0.1 ml/ 300 g Bdy Wt in canulae

## **Heparinized saline**

2 ml of 10000 Units/ml into 125 ml saline

## Endo Solutions

---

**Ketamine/Xylazine** (0.2 ml (of 87 mg ketamine and 13 mg xylazine per ml)

First 0.1 ml IP and a second 0,1 ml IM after the animal is down.

**a-Chlorolose** (56 mg/kg body weight) and **Urethane** (56 mg/kg body weight)

Initial dose is 0.1 ml/100 g Bdy Wt in canulae or 0.3 lm/300 gm Bdy Wt.

Maintenance dose 0.1 ml/ 300 g Bdy Wt in canulae

---

## Surgery

The rats will have their femoral artery cannulated following anesthesia with Ketamine/Xylazine (0.1 ml of 87 mg ketamine and 13 mg xylazine per ml per 100 g body weight). The femorals are exposed ventrally and cannulated. The incisions are kept moist with saline soaked gauze. Following surgery the rats are continuously observed by the TAs and Carl until the lab ends (approximately 3-4 hours post surgery) and then are euthanized by CO<sub>2</sub> inhalation. During the lab the students will take blood samples (0.5ml) at 10 minute intervals for approximately 2 hours under the direct observation of the TAs. Students will be monitored directly by Carl or the TAs. The depth of anesthesia will be monitored and corrected as needed. Following euthanasia via CO<sub>2</sub> inhalation, tissues will be harvested and the carcasses placed in a freezer for disposal.