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## Field anaesthetic and surgical techniques for implantation of intraperitoneal radio transmitters in Eurasian beavers *Castor fiber*

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Radio transmitters were implanted intraperitoneally in 22 (nine females, 13 males) adult, territorial Eurasian beavers Castor fiber under field conditions. Two different injectable anaesthestic drug combinations were tested. Access to the peritoneal cavity was made through a ventral midline incision. The animals in group # 1 (N = 10) were initially injected with medetomidine (0.05 mg/kg), ketamine (5 mg/kg) and butorphanol (0.1 mg/kg). Three animals needed additional injections of the drug combination. Muscle relaxation was poor and variable and some of the animals were sound sensitive. When midazolam (0.25 mg/kg) was added to the drug combination (group # 2), muscle relaxation was excellent and the beavers (N = 12) did not react to sound stimuli. All surgeries were successfully performed. One animal in group #1 died postoperatively due to circulatory failure. The behaviour and movements of the beavers did not appear to be affected by the procedure or the implant, except for the first few days when more time was spent inside the lodges. All beavers stayed in their original territory until they died, or as long as 17-24 months after the implantation. Based on these results, it appears that an injectable drug combination based on medetomidine, ketamine, butorphanol and midazolam and a surgical access through the ventral midline is suitable for implanting radio transmitters intraperitoneally in beavers under field conditions.

Key words: anaesthesia, Castor canadensis, Castor fiber, intraperitoneal implantation, radio-telemetry, surgery

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Implantation of radio transmitters intraperitoneally has been done in many wild beaver Castor spp. populations (Davis, Von Recum, Smith & Guynn 1984, Guynn, Davis & Von Recum 1987, Eisele, Faith, Menth, Parker & Van Vuren 1997, Nolet, Broekhuizen, Dorrestein & Rienks 1997, Wheatley 1997). Most researchers have used ketamine in combination with acepromazine or xylazine for anaesthesia (Gilbert & Gofton 1982, Wheatley 1997), but the quality of the anaesthesia and the total drug requirement(s) have not been well described. Some groups that have not been limited by field conditions have used volatile agents such as halothane and isoflurane (Greene, Keegan, Gallagher, Alexander & Harari 1991, Eisele et al. 1997). Surgical techniques previously described for intraperitoneal implantation of radio transmitters in beavers have been muscle split access trough a ventral-lateral approach (Wheatley 1997) or a paralumbar incision (Guynn et al. 1987). The aim of our study was to establish a protocol for surgical anaesthesia in Eurasian beaver Castor fiber under field conditions. We also wanted to evaluate the ventral midline incision as the surgical approach to the peritoneal cavity as this method offers the fastest and least traumatic access to the abdominal cavity in most animals.

## Material and methods

#### Study site and capture

The study was conducted on the Lunde (59° 17'N, 09° 06'E) and Gvarv (59° 25'N, 09° 04'E) rivers in the municipalities of Nome and Sauherad in the county of Telemark, Norway. We captured 22 adult, territorial Eurasian beavers (13 males and nine females; all > 4 years old) using landing nets (N = 12 territories; Rosell & Hovde 2001; Table 1). All results are presented as means ( $\pm$  SD). Beavers were transported ashore in a cloth sack and weighed. All beavers had previously been marked with coloured ear tags and a microchip in the neck, and had been sexed based on the colour of the anal gland secretion (Rosell & Hovde 2001).

#### Anaesthesia and surgery

When on land, the first 17 beavers were immediately injected with the anaesthetic drugs. The remaining six

animals (including a recapture, see Table 1) were transferred to a wooden box (1.0 x 0.5 x 0.7 m) and transported to the research facility where they were kept in a dark, quiet room until the next morning. Implantation of radio transmitters was conducted in 22 different beavers which had a mean body weight of  $22.3 (\pm 2.1)$ kg. One beaver had two implantations due to failure of the first radio transmitter implanted. The anaesthetic protocol varied, and the beavers were divided into two groups. In the first group (N = 10), the animals were anaesthetised with 0.05 mg medetomidine/kg (Domitor®) 1 mg/ml, Orion Corporation Animal Health, Turku, Finland), 5 mg ketamine/kg (Narketan® 100 mg/ml, Chassot AG, Bern, Switzerland) and 0.1 mg butorphanol/kg (Torbugesic® 10 mg/ml, Fort Dodge Laboratories, Fort Dodge, Iowa, USA). All drugs were injected intramuscularly in the gluteal muscle at the same time using separate syringes. In group #2 (N = 13), the anaesthetic protocol differed in the following way: In addition to the drugs used in group # 1, group # 2 was also injected intramuscularly with 0.25 mg midazolam/kg (Midazolam<sup>®</sup> 5 mg/kg, Alpharma AS, Oslo, Norway) which was injected at the same time as the other drugs. All animals were left undisturbed in the cloth sack or in the box until induction of anaesthesia was complete.

After induction of anaesthesia, the animals were placed in dorsal recumbency, and an area of 10 x 5 cm caudal to the umbilicus was clipped and swabbed with chorhexidine in 60% ethyl alcohol (Klorhexidin 5 mg/ml, Galderma Svenska AB, Bromma, Sweden). The underfur was not removed. Using standard surgical procedures (Fossum 1997), an approximately 6 cm long ventral midline incision was made to gain access to the peritoneal cavity. All animals were implanted with an Alterra® TX30.3A1 (egg-style; 65 x 34 mm) intraperitoneal 30 MHz radio transmitter (mean weight: 73 ( $\pm$  4) g) equipped with temperature and movement sensors (Alterra (IBN/DLO), NE-6700 AA Wageningen, the Netherlands). Estimated battery life time was 1,800 days. The radio transmitters were soaked in 10 mg/ml benzalkonium chloride, prewarmed to body temperature and rinsed with sterile saline before being placed to float freely in the peritoneal cavity. The incision was closed in two layers using a Polydioxanone 1-0 absorbable

Table 1. Names, capturing dates, sex, weight (in kg), drug treatment and follow up/fate of the 22 Eurasian beavers surgically implanted with radio transmitters in the municipality of Bø, Norway. The drug combinations were: 1) medetomidine 0,05 mg/kg, ketamine 5 mg/kg, butorphanol 0,1 mg/kg, and 2) medetomidine 0,05 mg/kg, ketamine 5 mg/kg, butorphanol 0,1 mg/kg, midazolam 0,25 mg/kg).

Name	Capture date	Colony	Sex	Weight	Drug combination	Follow up/ Fate
Jon	18.09.99	L1	đ	22.0	1	Alive in September 2001
Trude	18.09.99	L1	ę	22.5	1	Alive in September 2001
Ørjan	18.09.99	L2a	đ	24.0	1	Alive in September 2001
Ørjan <sup>1</sup>	05.04.00	L2a	đ	24.0	2	Alive in September 2001
Birgit <sup>4</sup>	18.09.99	L2a	Ŷ	24.0	1	Alive in September 2001
Frode	04.04.00	L2b	đ	20.0	2	Alive in September 2001
Blondi <sup>2</sup>	04.04.00	L2b	Ŷ	23.0	2	Alive in September 2001
Grønn	04.04.00	L3	ð	20.0	2	Alive in September 2001
Jørn	18.09.99	L4	ð	22.0	1	Alive in September 2001
Hanna	18.09.99	L4	Ŷ	23.5	1	Alive in September 2001
Bjørk <sup>2, 4</sup>	03.04.00	L5	Ŷ	21.5	2	Contact lost in December 2000 <sup>3</sup>
Sonja <sup>2, 4</sup>	03.04.00	L6	Ŷ	23.0	2	Alive in September 2001
Harald	04.04.00	L6	đ	21.5	2	Alive in September 2001
Greg Burly	02.05.00	Haugen	đ	17.5	2	Alive in September 2001
Jan Helge	19.09.99	Evju	đ	22.5	1	Died post-operatively in 2001 <sup>3</sup>
Frøydis	19.09.99	Evju	Ŷ	24.5	1	Found dead in May 2000 <sup>3</sup>
Marcus	02.05.00	Evju	đ	21.0	2	Found dead in July 2000 <sup>3</sup>
Lt. Kristiansen	02.05.00	Gvarv	đ	21.0	2	Alive in September 2001
Tåkehode	19.09.99	N1	đ	25.0	1	Alive in September 2001
Sofie	19.09.99	N1	Ŷ	26.5	1	Shot in July $2001^3$
Jobu	02.05.00	N1	đ	18.0	2	Found dead in January 2001 <sup>3</sup>
Hr. Nilsson	02.05.00	N2	đ	20.0	2	Alive in September 2001
Pernille <sup>2, 4</sup>	25.04.00	N2	Q	24.0	2	Alive in September 2001

<sup>1</sup> First radio transmitter faulty, new radio transmitter implanted.

<sup>2</sup> Pregnant.

<sup>3</sup> Jan Helge died within three hours after surgery was completed. Bjørk probably died or dispersed (was not found) because another adult female moved into her territory. Frøydis probably died due to old age (estimated to be 20 years). Marcus died due to a collapsed burrow. Sofie was illegally shot. Jobu probably died due to under- or malnutrition (one damaged incisor in both the upper and lower jaw.

<sup>4</sup> Gave birth the following summer.

suture (Ethicon PDS II, Johnson & Johnson Intl.). Simple, interrupted sutures were placed in the *linea alba*, and a continuous, horizontal mattress suture in the skin. During surgery, abnormal extension and rigor of limbs and neck indicated poor muscle relaxation. Obvious reaction when surgical instruments were put in metal case was used as an indication of increased sound sensitivity. This was subjectively assessed.

A pulse oximeter (NELLCOR® N-20P, Nellcor Inc., Pleasanton, CA, USA) sensor (VetSat®, Nellcor Inc.) was attached to the tongue, and the relative arterial oxygen saturation  $(SpO_2)$  and pulse rates were monitored. If  $SpO_2$  dropped below 80% in group # 2, the animals were allowed to breathe 100% oxygen through a face mask. After surgery, the animals were injected intramuscularly with procaine/benzathine penicillin 100.000 IU/kg (Penikel® 15+15, Kela Laboratoria NV, Hoogstraten, Belgium) to minimise the risk of postoperative wound infections. To shorten the recoveries from anaesthesia, all animals were injected intramuscularly with 0.25 mg/kg atipamezole (Antisedan® 5 mg/ml, Orion Corporation Animal Health). The induction time was defined as the time interval from injection of the drugs until the animals became recumbent in the box and could be handled without making resistance. Surgical time was defined as the interval from the skin incision until the skin suture was finished. The beavers were kept in a wooden box in a quiet and dark area until recovery, after which they were released into the water at the site where they had been captured. The time from injection of atipamezole until recovery was not recorded because postoperative observations were impeded by the animals being allowed to recover in a dark, quiet box in order to minimise additional stress.

#### Follow up of animals

After surgery, beavers were radio tracked daily during the first week, then weekly for the next two months and thereafter monthly for a year. The study was approved by the Norwegian Animal Research Authority.

## Results

#### Anaesthesia and surgery

In group # 1, the mean induction time was 8.4 ( $\pm$  5.0) minutes, and the mean surgical time for implantation of the radio transmitter was 26 ( $\pm$  10) minutes. Induction of anaesthesia was generally calm; muscle relaxation was variable and was considered to be poor in three animals. Three of the beavers were also very sound sensitive. Two animals needed one top-up dose of the anaesthetic

drugs to achieve surgical anaesthesia. Top-up doses were calculated as half the initial dose of medetomidine, ketamine and butorphanol. During surgery, mean SpO<sub>2</sub> was 78 ( $\pm$  9)%, and mean pulse rate was 54 ( $\pm$  4) beats/ minute. All animals in group # 1 had episodes when SpO<sub>2</sub> dropped below 80%.

In group # 2, the mean induction time was 7.8 ( $\pm$  2.6) minutes, and the mean surgery time was 15.6 ( $\pm$  6.1) minutes. Induction was calm, muscle relaxation was good throughout the procedure and the beavers did not seem to react to sound stimuli. Mean SpO<sub>2</sub> was 84 ( $\pm$  4)% and mean pulse rate during surgery was 54 ( $\pm$  7) beats/minute. Brief episodes when SpO<sub>2</sub> dropped below 80% occurred in seven animals, which were then allowed to breathe 100% oxygen through a face mask held over the animals' noses.

All surgeries were successfully performed, and no specific problems were encountered neither during surgery, in the postoperative period nor before the beavers were released back into the water at the site of capture.

#### Survival

In group # 1, one animal did not recover from anaesthesia and was found dead in the box approximately three hours after surgery (see Table 1). An autopsy performed at the National Veterinary Institute, Oslo, Norway, led to the conclusion that circulatory failure was the likely cause of death. No signs of wound dehiscence were found, and the implanted transmitter was floating freely within the peritoneal cavity.

The behaviour and movements of the beavers did not appear to be affected by the procedure or implants, except for the first few days when they spent more time inside their lodges. All beavers (fate of one beaver is unknown) stayed in their original territory until they died (four animals) or as long as 17-24 months after implantation (see Table 1).

## Discussion

The anaesthetic regime used in group # 1 (i.e. medetomidine, ketamine, butorphanol) did not provide a reliable anaesthesia, because the side effects were not acceptable. This was clearly evident from the additional injections that had to be made in three animals, and from the muscle rigor and sound sensitivity displayed in some of the beavers. Ketamine combined with acepromazine was used by Wheatley (1997) to anaesthetise North American beavers *C. canadensis* during implantation of radio transmitters. In that study, repeated injections of ketamine were given to maintain anaesthesia, and the beavers' legs were tied to inhibit 'involuntary movement', an indication of inadequate anaesthesia during surgery.

Because of inadequate anaesthesia in group # 1, the anaesthetic regime was modified. In other species, ketamine has been combined with benzodiazepines to increase muscle relaxation and decrease CNS responsiveness (Hall, Clarke & Trim 2000). Midazolam is a water soluble benzodiazepine with a rapid effect and little tissue toxicity, and is therefore suitable for intramuscular injection. We also felt the need to increase the SpO<sub>2</sub> without having to intubate the animals with an endotracheal tube. Therefore, animals in group # 2 were allowed to breathe 100% oxygen through a face mask when the relative oxygen saturation dropped below 80%.

None of the animals in group # 2 needed additional injections of the anaesthetic drugs, and muscle relaxation was good. Despite the midazolam respiratory depressant properties, the mean  $SpO_2$  was higher in group # 2 than in group # 1. This is probably attributable to the supplementation of pure oxygen in group # 2. The shorter surgical time in group # 2 may also indicate that the anaesthetic regime improved when midazolam was added.

The normal heart rate in beavers is reported to be 120 beats/minute when animals are active (e.g. grooming or swimming; Swain, Gilbert & Robinette 1988). Diving causes a bradycardia (61 beats/minute), and beavers also show a 'passive defence response' when threatened on land. In such cases, the behavioural responses are accompanied by a marked bradycardia which is of the same magnitude as the diving bradycardia (Swain et al. 1988). All beavers had low pulse rates during anaesthesia in both groups. Whether the anaesthetic drugs caused this bradycardia *per se*, or whether the anaesthesia induced a diving reflex or a 'passive defence response', could not be determined.

We chose the ventral midline access because this approach offers the fastest and least traumatic access to the abdominal cavity in most mammals. Other researchers have used a ventral-lateral incision (Wheatley 1997) or a paralumbar incision in the beaver (Davis et al. 1984), and they argued that due to the anatomy and the behaviour of beavers, more abrasion of the incision would occur if ventral midline incision was used. In our opinion, a muscle-split technique as described by Wheatley (1997) or Davis et al. (1984) will cause more bleeding, last longer, increase the risk of postoperative infections and most probably be more painful in the postoperative period due to the greater extent of tissue trauma. In 22 North American river otters *Lontra canadensis*, the

access to the abdominal cavity was made through the paralumbar fossa (Hernandez-Divers, Kollias, Abou-Madi & Hartrup 2001). In two of these animals, serious complications occurred, i.e. haemorrhage and postoperative infection. In addition, there may be a risk of injuring the nerves to the abdominal wall by using a lateral incision. The ventral midline approach has been used in more than 400 surgeries on free-ranging brown bears Ursus arctos, lynx Lynx lynx, wolverines Gulo gulo, European river otters Lutra lutra and European wild boars Sus scrofa in Scandinavia (Arnemo 1991, Arnemo, Dypsund, Berntsen, Schultze, Wedul, Ranheim & Lundstein 1998, Arnemo, Linnell, Wedul, Ranheim, Odden & Andersen 1999). In all these studies, two animals have been found dead due to wound dehiscence. If the short surgical wound in the linea alba is closed with a sufficient number of sutures and the knots are adequately secured, it is unlikely that the wound will break open or that a hernia will occur. The use of the polydioxanone (PDS) monofilament suture causes less inflammation and maintains tensile strength longer than other absorbable sutures (Sanz, Patterson, Kamath, Willett, Ahmed & Butterfield 1988).

In summary, the anaesthetic combination of medetomidine, ketamine, butorphanol and midazolam administered by intramuscular injection is well suited for surgical implantation of radio transmitters in beavers under field conditions. The ventral midline approach to the peritoneal cavity proved to be fast, simple and without postoperative complications.

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