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Use of intraperitoneal radio-transmitters in lynx *Lynx lynx* kittens: anaesthesia, surgery and behaviour

Jon M. Arnemo, John D.C. Linnell, Sari J. Wedul, Birgit Ranheim, John Odden & Reidar Andersen

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The suitability of intraperitoneally implanted radio-transmitters as a method for studying young lynx *Lynx lynx* kittens was tested under field conditions. Radio-marked adult females were followed to the lair where they kept their kittens. In 1997 and 1998, nine kittens (4-5 weeks old) were located. One of two implant models (7 g and 20 g) were implanted using surgical procedures and a combination of medetomidine (0.08 mg/kg) and ketamine (5 mg/kg) for anaesthesia. No complications occurred during the operations. All kittens were accepted again by their mother and were moved to a new lair within 1 km. All survived at least three months after the operation. Six of the kittens were re-examined 4-5 months after the operation. In all of these cases the implants were floating freely in the peritoneal cavity. Based on these results it appears that intraperitoneal implanting of radio-transmitters is a very useful method for studying very young lynx kittens, and could be used for most felids of a similar, or larger, size.

Key words: anaesthesia, intraperitoneal implantation, lynx, *Lynx lynx*, radio-telemetry, radio-transmitters, surgery

Jon M. Arnemo, Department of Arctic Veterinary Medicine, Norwegian School of Veterinary Science, N-9292 Tromsø, Norway & Department of Forestry and Wilderness Management, Hedmark College, Evenstad, N-2480 Koppang, Norway

John D.C. Linnell, Norwegian Institute for Nature Research, Tungasletta 2, N-7485 Trondheim, Norway - e-mail: john.linnell@ninatrd.ninaniku.no

Sari J. Wedul, Elverum Animal Clinic, N-2400 Elverum, Norway

Birgit Ranheim, Department of Pharmacology, Microbiology and Food Hygiene, Norwegian School of Veterinary Science, PO Box 8146 Dep., N-0033 Oslo, Norway

John Odden, Department of Forestry and Wilderness Management, Hedmark College, Evenstad, N-2480 Koppang, Norway

Reidar Andersen, Norwegian Institute for Nature Research, Tungasletta 2, N-7485 Trondheim, Norway & Zoology Department, Norwegian University of Science and Technology, N-7055 Dragvoll, Norway

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The scientific study of wild carnivore behaviour, ecology and population dynamics is virtually dependent on radio-telemetry (Samuel & Fuller 1996). Without the use of telemetry, estimates of survival, reproduction, and predation behaviour can be severely biased. While methods for the capture and marking of adult-sized animals are well established (Poole, Mowat & Slough 1993, Mowat, Slough & Rivard 1994, Samuel & Fuller 1996, Nybakk, Kjørstad, Overskaug, Kvam, Linnell, Andersen & Berntsen 1996), those for neonates and juveniles are few. This lack of information is especially important when it is considered that juvenile mortality is important in shaping population dynamics and life history strategy (Promislow & Harvey 1990, Laurenson 1994). Litter size at birth is usually known from necropsy data (e.g. Kvam 1991), but data on mortality and behaviour during the first six months of life are lacking.

Attempts have been made to develop expanding collar or harness systems for juvenile carnivores (Garcelon 1977, Strathearn, Lotimer & Kolenosky 1984, Jackson, Jackson & Seitz 1985, Blackwell, Frost, Flinders & Barber 1991, Robertson & Harris 1996), but these have not been as successful as those used on juvenile ungulates (Linnell, Aanes & Andersen 1995). They have all had problems in that they either have a short expansion range and therefore they are capable of providing data only for a short period of the juvenile's development (Robertson & Harris 1996), or they can only be fitted to large juveniles that have left the lair or den stage of development (Garcelon 1977), or they fall off prematurely (Weber & Meia 1992, Fredrickson & Mack 1996). Attempts to glue small transmitters into the fur of 4-week-old lynx kittens failed, because the mother removed the transmitters within 24 hours (J.D.C. Linnell, R. Andersen & P. Segerström, unpubl. data).

Implanting radio-transmitters into the peritoneal cavity has been used for over 20 years on adult carnivores (e.g. Melquist & Hornocker 1979, Reid, Melquist, Woolington & Noll 1986, Arnemo 1991, Samuel & Fuller 1996), but as far as we know it has only been used in a few studies on juveniles, e.g. bobcats *Lynx rufus* (Fredrickson, Mack, Gertonson & Stangle 1993), coyotes *Canis latrans* (Green, Gollightly, Lindsey & LeaMaster 1985, Gese, Ruff & Crabtree 1996), wolverines *Gulo gulo* (Copeland 1996, Arnemo, Dypsund, Berntsen, Schulze, Wedul, Ranheim & Lundstein 1998a) and brown bears *Ursus arctos* (Arnemo et al. 1998a). In this paper we describe the successful application of implant transmitters

to neonatal (<5 weeks old) Eurasian lynx *Lynx lynx* kittens in a field study in Norway.

Material and methods

Capture

The date of birth for each radio-collared female lynx was estimated from changes in their movement pattern during May-June. Each female decreased her daily movements and adopted a central-place foraging pattern at the assumed time of birth (Schmidt 1998). When the kittens were estimated to be 4-5 weeks old the adult female was monitored on a hunting excursion away from the lair. Upon her return, she was closely stalked by one or two people with radio-tracking equipment and approached to within 50 m and used as a cue for locating the kittens. At this age the kittens would either remain lying on the ground, or crawl beneath some stones or dense brush. In either case, they could be picked up and handled without the use of any restraint. Once the two stalkers had located the kittens the rest of the field crew (2-6) were allowed to approach.

Anaesthesia and surgery

Surgery was performed in the field immediately after capture. Anaesthesia was induced with a combination of medetomidine (Domitor® 1 mg/ml, Orion Corporation Animal Health) and ketamine (Ketalar® 50 mg/ml, Parke-Davis). Kittens were manually restrained, weighed and injected intramuscularly with 0.08 mg/kg of medetomidine and 5 mg/kg of ketamine from the same syringe. After drug administration, the kittens were left undisturbed in a dark bag during induction of anaesthesia.

Kittens were kept in dorsal recumbency during the entire surgical procedure. A pulse oximeter (NELLCOR® N-20P, Nellcor) with the sensor (DURA-Y®, Nellcor) attached to the tongue was used to monitor relative arterial oxygen saturation and pulse rate. Rectal temperature (using a digital clinical thermometer) was recorded at irregular intervals. An eye gel (Viscotears® CIBA Vision) was used to protect the cornea.

Intraperitoneal implantation of radio-transmitters (Telonics®, CHP-IMP, 7 g, 3.3 x 1.9 x 1.0 cm, 6 months battery life at 40 pulses/minute, or IMP/150/L, 20 g, 5.3 x 2.3 cm, 6 months battery life at 40 pulses/minute) was performed using standard surgical procedures (Fossum 1997). Hair in an area of 2 x 5 cm caudal to the umbilicus was clipped and lightly swabbed

with an antiseptic (chlorhexidine and ethyl alcohol, Klorhexidin® 5 mg/ml, Nycomed Pharma). A ventral midline approach for access to the peritoneal cavity was used (Fossum 1997). After soaking in 10 mg/ml benzalkonium chloride for one hour, prewarming to body temperature and washing with sterile saline, the transmitter was placed freely within the peritoneal cavity. The incision was closed in two layers using 2-0 polyglactin 910 absorbable suture (Vicryl®, Ethicon) in a simple interrupted pattern for the *linea alba* and in an interrupted horizontal mattress pattern for the skin (Fossum 1997).

Following surgery, kittens were injected intramuscularly with 25 mg/kg of dihydrostreptomycin and 20 mg/kg of procaine benzylpenicillin (Streptocillin® Boehringer Ingelheim Agrovvet) in order to reduce the potential risk of infection. All kittens were tagged subcutaneously with a microchip (INDEXel®, Rhône Mérieux) on the left side of the neck. For reversal of anaesthesia, kittens were injected intramuscularly with 0.16 mg/kg of atipamezole (Antisedan® 5 mg/ml, Orion Corporation) and were placed in a dark bag. When recovery was complete, kittens were returned to the lair.

Efforts were made to recapture living kittens during the following autumn in order to change the implant before the batteries expired. Hunting dogs were used to chase the animals into a tree, and 0.2 mg/kg of medetomidine (Zalopine® 10 mg/ml, Orion Corporation Animal Health) and 5 mg/kg of ketamine (Ketalar® 50 mg/ml) were administered with a tranquillising gun (Daninject®), 3 ml plastic darts, with 1.5 x 30 mm collared needles. For safety, a large net was stretched under the tree in case the animal fell after darting. Following the same surgical procedures

as in kittens, the smaller implant was substituted with a new transmitter (Telonics® IMP/400/L, 90 grams, 9.7 x 3.3 cm, 36 months battery life at 40 pulses/minute). All juveniles were injected intramuscularly with 0.4 mg/kg of atipamezole for reversal of anaesthesia, and were observed until they were able to walk away.

Behavioural responses to marking

The responses of mothers during both the tagging procedure and after the kittens were returned to the lair, were monitored using radio-telemetry. We recorded maximum distance away from the lair during marking, the time elapsed before their return, and the distance which they moved the kittens to a new lair following their return.

Results

Capture

Four litters were handled in 1997 and two in 1998. The kittens were between four and five weeks of age (Table 1). Although only six kittens were found in the four litters of 1997, later observations during autumn revealed that all litters had contained two kittens. The stones and vegetation at the lairs could easily have hidden the other kittens, although it is also possible that the litter was split in two locations. Three kittens were found in the two 1998 litters, and subsequent observations did not indicate the presence of additional litter-mates. Five of the litters were produced by pluriparous females, whereas the sixth was produced by a 3-year-old primiparous female.

Table 1. Behavioural responses of female lynx to the disturbance caused by implanting intraperitoneal transmitters in their neonatal kittens.

| Mother | Kittens | Estimated birth date | Marking date | Mother's flight distance | Delay before return (hrs: min) | Distance moved to new lair (km) | Period monitored (days) ³ | Follow-up / Fate |
|-------------------|---------|----------------------|--------------|--------------------------|--------------------------------|---------------------------------|--------------------------------------|-------------------------------|
| #121 | 124 (♂) | 26.05.97 | 03.07.97 | ≈2 km | 6:45 | 0.7 | 121 | Lost contact, early 11.97. |
| | 125 (♂) | | 03.07.97 | | | | 122 | Implant changed 01.11.97 |
| #111 | 126 (♂) | 05.06.97 | 03.07.97 | ≈1 km | 11:15 | 0.1 | 118 | Implant changed 28.10.97 |
| #119 | 127 (♀) | 06.06.97 | 08.07.97 | ≈2 km | < 24 hrs ² | 0.9 | 119 | Implant changed 03.11.97 |
| #107 ¹ | 128 (♂) | 07.06.97 | 15.07.97 | ≈5 km | 19:45 | 0.5 | 108 | Died of kachexia 30.10.97 |
| | 129 (♀) | | 15.07.97 | | | | 108 | Died of kachexia 30.10.97 |
| #111 | 135 (♀) | 26.05.98 | 02.07.98 | ≈1 km | 0:45 | 1.7 | 152 | Survived at least until 12.98 |
| | 136 (♂) | | 02.07.98 | | | | 152 | Survived at least until 12.98 |
| #121 | 137 (♀) | 26.05.98 | 08.07.98 | ? | < 24 hrs ² | 2.0 | 92 | Implant changed 07.10.98 |
| Means | | 27.05 | 07.07 | 2.7 | 9:37 | 1.0 | 121 | |

¹ This reproduction was by a primiparous female, the other eight reproductions were by pluriparous females.

² For two females the exact delay was not recorded, but they had returned when the animals were checked the next day.

³ The period monitored describes the period up until the implant was changed, the batteries stopped or the kitten died.

Anaesthesia and surgery

The mean body mass of kittens was 1.45 kg (SD = 0.27, range: 0.90-1.75). Inductions were rapid and calm in all kittens. The mean time from injection of medetomidine-ketamine to complete immobilisation was 2.5 minutes (SD = 1.1, range: 1.0-4.0).

Muscle relaxation was excellent and there were no symptoms of pain or spontaneous movements during surgery. No complications were encountered and surgeries were performed in less than 20 minutes. Two kittens received Telonics® CHP-IMP while seven were implanted with Telonics® IMP/150/L. The mean time from injection of medetomidine-ketamine to administration of atipamezole was 36 minutes (SD = 6, range: 25-41; N = 8).

Mean rectal temperatures recorded immediately after induction and before administration of atipamezole were 37.8°C (SD = 1.4, range: 36.7-38.9; N = 6) and 37.0°C (SD = 1.6, range: 35.9-40.0; N = 6), respectively. Means (N = 5) for pulse rate (beats/minute) and relative arterial oxygen saturation (%) were 148 (SD = 25, range: 78-192) and 93 (SD = 5, range: 80-100), respectively.

Effective reversals followed administration of the atipamezole, mean time from injection of the antagonist to when the kittens were awake and able to move was 4.8 minutes (SD = 4.2, range: 2-13 minutes). All kittens were alert and calm, and excitement or other signs of abnormal response were not seen. All kittens were returned to the lair within 30 minutes post surgery.

Four juveniles with a mean body mass of 6.4 kg (SD = 0.9, range: 5.6-7.3) were recaptured 92-122 days (mean = 112 days) after the first implantation. After induction of anaesthesia, a person climbed the tree to bring down the immobilised animals. No clinical complications were encountered during surgeries. The previous implants were floating freely in the peritoneal cavity and were easily located and removed. No adhesions or other side effects from the first surgery were visible.

Behavioural responses to marking

In all cases mothers immediately ran away from their kittens when approached, and never showed any signs of aggression towards the approaching stalkers. During the operations mothers generally circled the lair within 100 m before moving away a few kilometres (see Table 1). The movement of 5 km made by the primiparous female appeared to be extreme.

In every case the mothers returned to the kittens within 24 hours (see Table 1) and accepted them. No

attempt had been made to minimise the amount of human scent transferred to the kittens. The primiparous female had the longest delay before returning. The kittens were immediately moved to a new lair, within 1 km of the first, following the mother's return (see Table 1).

No immediate mortality resulted from the surgery. All the implanted kittens survived at least three months after the operation. The two kittens belonging to the primiparous female were found dead 3.5 months after implantation. Necropsy revealed kachexia, heavy nematode infestations and moderate occurrence of sarcoptic mange mites in both. The wounds from the surgery were completely healed and no reactions to the implant were noted in the post-mortem examinations.

Discussion

Medetomidine-ketamine was an excellent combination for surgical anaesthesia in neonatal lynx. Inductions were rapid and calm, muscle relaxation was good, and there was no pain reaction to laparotomy. The dose levels used were similar to those recommended for domestic cats (Flecknell 1994, Arnemo, Ranheim & Sjøli 1998b). Although mild hypoxemia, defined as a relative arterial oxygen saturation <90 % (Trim 1994), was seen in two kittens, side effects of clinical significance were not seen. Recoveries after atipamezole administration were smooth and rapid in all kittens and post-surgical complications were absent. During recapture of juveniles, a higher dose of medetomidine was used in order to ensure rapid immobilisation. In other carnivore species it is well established that stress and excitement increase the effective dose of immobilising drugs (Arnemo et al. 1998b). In general, the weight of the implant should not exceed 2% of the body mass of the animal (Arnemo et al. 1998a). In our study, the weight of the Telonics® IMP/150/L was less than 1.5% of the smallest kitten's body weight.

Access to the peritoneal cavity was by a ventral midline approach (Fossum 1997). In some studies using intraperitoneal implants in wildlife (e.g. Melquist & Hornocker 1979, Serfass, Peper, Whary & Brooks 1993) a lateral or flank approach has been applied. However, this method causes more bleeding and tissue damage and is, therefore, more difficult to perform and more painful to the animal during the post-operative period, although it may be more appropriate for species, like otters *Lutra* sp., that often drag

their ventral surface on the ground (Serfass et al. 1993). The ventral midline approach has been used in more than 160 surgeries on free-ranging brown bears, lynx and wolverines in Scandinavia, and is recommended for similar studies of felids (Arnemo et al. 1998a).

All kittens and juveniles with implants had apparently normal behaviour in our study, and seven out of nine animals lived for the life expectancy of the battery (5-7 months) after the first implantation. We could not evaluate possible long-term effects, but necropsies of the two kittens that died 3.5 months after surgery, showed complete healing without any signs of reaction to the implant. Also, in the four juveniles that underwent a second surgery in order to change the implant, no side-effects from the first operation or from the implant were noted.

All the mothers accepted their kittens after surgery and only moved them a short distance to a new lair. These results were encouraging as there are still very little data about the degree to which carnivores tolerate human disturbance at their natal dens or lairs (Copeland 1996, Thiel, Merrill & Mech 1998), or handling of their young. Among ungulates, marking-induced abandonment of fawns is an important concern (Livezey 1990) which necessitates careful procedures. It appeared that lynx mothers tolerated our activities, which is important if such invasive methods become widespread. The delay in returning to the kittens and their subsequent relocation may have exposed them to some degree of additional mortality risk, however at this age the mother generally spends at least 12 hours of each day hunting up to 10 km away from the kittens (J.D.C. Linnell & R. Andersen, unpubl. data, Schmidt 1998). Therefore, it was likely that the kittens were able to thermoregulate without their mother, and that the lair provided enough physical protection from the elements and potential predators.

In conclusion, we recommend the use of intraperitoneal transmitters for radio-tagging neonatal and juvenile Eurasian lynx, and the method can most probably be applied to other felids of similar size. There were no observable negative effects of implanting on neonatal kittens, and we demonstrate that marked individuals can be recaptured to change, or remove, the implants if required.

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