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RESEARCH

A field test of the anaesthetics, surgical methods and radio-transmitters required for producing Judas pigs for an eradication programme

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Abstract: Judas pigs (*Sus scrofa*) will be integral to the success of the proposed Auckland Island (45 891 ha) pig eradication. Judas pigs must be permanently unable to breed and reliably retain a radio-transmitter to enable tracking and retrieval. This study tested the techniques and practical requirements to undertake the procedures required to produce a Judas pig in a remote location. Four adult female wild pigs were captured using a net-gun from a helicopter and processed at a base site in the field. They were anaesthetised using intramuscular injection of one of two anaesthetic protocols (Zoletil™, ketamine and xylazine; or medetomidine, ketamine and butorphanol). Surgical sterilisation consisted of tubal ligation and resection via a ventral midline incision. Pigs received either a subcutaneous or intra-abdominal implanted radio-transmitter and had a radio-collar and radio-ear tag attached. The anaesthetic was reversed prior to release close to the capture location. Radio-tracking at 10 and 104 days showed all pigs had survived and radio-tracking was effective using all the transmitter types. Pigs were humanely dispatched and necropsied on day 104. No adverse events were detected from the surgeries or radio-transmitters. All transmitter types except one subcutaneous transmitter model were shown to be secure and had an effective radio range for a Judas pig programme. Pig growth rates implicate a limited humane collaring period and ear transmitters may be at greater risk of transmitter loss. Implantable intra-abdominal radio transmitters provide the most security, but multiple transmitters are advised. Surgical sterilisation by tubal ligation and resection under general anaesthesia using an intramuscular injection containing medetomidine, butorphanol and ketamine and reversed with atipamezole was shown to be feasible in field conditions.

Keywords: anaesthesia, implantation, Judas pig, radio-transmitter, sterilisation, surgery, *Sus scrofa*

Introduction

The Auckland Islands (56 816 ha), in the New Zealand subantarctic islands lying 465 km south of mainland Aotearoa/New Zealand (hereafter NZ), are a Nature Reserve and World Heritage Site. The archipelago is a stronghold for taonga, including, but not limited to, several species of toroa (albatrosses, family Diomedeidae), tītī (petrels, family Procellariidae), hoiho (yellow-eyed penguin, *Megadyptes antipodes*), and whakahao/rāpoka (sealion, *Phocarctos hookeri*). Of the 38 native bird taxa, 25 are seabirds, which includes three endemic species (Miskelly et al. 2020). The largest island, Auckland Island (45 891 ha) is the only island in the archipelago, and the New Zealand subantarctic islands, with remaining populations of mammalian pest species: feral pigs (*Sus scrofa*), cats (*Felis catus*), and mice (*Mus musculus*).

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The Maukahuka Pest Free Auckland Island project aims to eradicate all three mammalian pest species. The planned removal of feral pigs from Auckland Island is one of the largest and most complex projects of its type in the world.

Removing feral pigs will rely on the systematic application of a range of eradication tools. In addition, detecting survivors over such a large landmass presents a significant challenge (Parkes et al. 2010). Conventionally used on goats (Campbell & Donlan 2005), the Judas technique involves individuals in a population that are fitted with a radio transmitter. Being social animals, these individuals group themselves with other non-Judas animals allowing practitioners guided by the radio transmitter to find and dispatch the previously undetected animals. Judas pigs are widely used to increase efficiencies of feral pig control (McIllroy & Gifford 1997; Wilcox et al. 2004; McCann & Garcelon 2008) and were also an integral tool in

the eradication of feral pigs from Santa Cruz Island (California, USA) in 2007 (Parkes et al. 2010). It is proposed that Judas pigs will provide a high-confidence, cost-effective method to detect any pigs that survive the eradication programme on Auckland Island.

The Auckland Island pig eradication faces additional challenges due to the island's size, thick scrub and challenging terrain (Challies 1975). The Judas programme is likely to extend over at least 12 months, which requires an extremely robust approach to reproductive management and transmitter security for the Judas pigs. It is integral to the Judas pig programme to prevent transmitter loss or failure leading to Judas pigs that cannot be tracked. Radio-ear tags and radio-collars, while successful in many circumstances, may not be suitable (or solely suitable) for this remote programme given the risk of becoming dislodged or damaged and the animal welfare issues relating to the growth of the pigs after collar attachment (Arnemo et al. 1999). Although external devices may have greater detectability (Coetsee et al. 2016), implantation of radio transmitters is a technique that offers long-term transmitter security (Ralls et al. 1989; Coetsee et al. 2016), which is considered essential for this eradication programme.

Many different options are available for radio-transmitter implantation techniques. Intra-abdominal implantation can be free-floating (Arnemo et al. 1999) or attached to or within the musculature (Willens et al. 2014). Subcutaneous implantation can be with or without an external aerial (Mulcahy & Garner 1999). Potential adverse animal welfare effects may occur with any implantation and include infection, interference with abdominal organs such as tissue adhesions, physical interference with reproductive capacity with intra-abdominal implants (Lechenne et al. 2012), subcutaneous fluid pockets and muco-purulent discharge due to tissue reaction to the implanted device (Lander et al. 2005; Horning et al. 2017), and breakdown of surgical closures due to interference by the target animal (Moons et al. 2007).

This trial's objective was to field test the techniques required to produce a Judas pig in a remote field situation. It explored the use of two anaesthetic protocols for field surgery, the surgical technique for permanent sterilisation of females, and intra-abdominal or subcutaneous implantation of radio transmitters. Assessment of security, ease of attachment, detectability and reliability of four transmitter types (external collar, external ear tag, implantable subcutaneous and free-floating intra-abdominal implant) were also investigated.

Judas pigs will have multiple encounters with humans during the eradication that is likely to induce avoidance behaviours. Surgical sterilisation is considered necessary to ensure irreversible life-time sterility to mitigate the risk of any Judas pigs not being retrieved at the end of the eradication (due to learned aversion or transmitter failure). Surgical sterilisation of females requires a deeper level of anaesthesia than routine sedation for handling. Injectable anaesthetic protocols were employed to allow abdominal surgery in a field situation. Males were not included in this study since surgical sterilisation of males is significantly less invasive and thus would not provide the same assessment of the practical complexities compared with female sterilisation. It is intended that both male and female pigs would be used in the eradication programme.

This study was a proof-of-concept approach, as all techniques have been previously successfully applied in pigs or other species. The aim was to test the logistics required to produce Judas pigs under field conditions and develop first-hand experience within the team to apply the lessons to the future eradication programme for feral pigs on Auckland Island.

Methods

Study site

The study was undertaken in the Takitimu Conservation Area east of Te Anau in Southland, South Island, New Zealand (45.74° S, 167.81° E; Fig. 1). Pigs were sampled from Telford and Gibraltar catchments, areas of known high feral pig densities and restricted public access to reduce the risk of recreational hunters capturing or killing the study pigs.

Capture and restraint

Pigs were located by helicopter in the open scrublands within a 5-min flight time from the surgical base with a maximum allowable chase for the capture of 5 mins to minimise stress and risk of capture myopathy or malignant hyperthermia in the pigs. Small to medium-sized female pigs (15–35 kg) were targeted to enable easy handling. Pigs were captured using a net gunshot from the helicopter, then a handler jumped out to restrain the pig in the net and apply hobbles. The fore and hind legs were strapped together in a flexed sitting position, using two wide soft leather belts, one each for the left and right sides. A black pillowcase with a drawstring was placed over the head and the drawstring lightly tightened around the neck to secure it. The pigs were transported directly to the operating site and weighed on arrival using a hanging stock scale.

Pigs were captured individually with a stand-down time between captures to enable the surgical procedure to be almost completed before the next pig was captured.

The pigs were left approximately 8 m from the surgery area until ready for surgery and protected from disturbance using a barrier, which also provided shade and protection from the wind. Blankets were used to cover the pigs when the temperature dropped below approximately 13°C.

Pigs were randomly assigned to each anaesthetic or surgical protocol based on the order of capture; deviation was allowed to adjust the allocation if required for animal welfare reasons.

Anaesthesia

All pigs received an external veterinary examination for general health, and the resting heart rate and respiratory rate were recorded prior to anaesthesia. The pigs were still hobbled and were further restrained by a handler applying pressure at the shoulder and hip. The injection site on the rump was cleaned with alcohol swabs and each pig was injected with anaesthetic drugs deep into the thigh muscle, with 40 mm × 18 or 19 gauge (G) needles. Eye protection, gloves, and Luer-lock syringes were used for safety precautions against syringe splash-back onto the operator. Once the pig was no longer responding to stimuli at approximately 5–10 mins after induction, the hood and restraints were removed. Each pig had a 25 mm x 22 G or 24 G intravenous (IV) catheter (BD Insyte™ IV Catheter, Becton, Dickinson and Company, USA) inserted into the lateral or medial auricular vein and secured with tape (Leukoplast™ Sleek tape, BSN Medical, Hamburg, Germany). Lacrilube™ (Allergan, Australia) was applied to the pigs' eyes to protect them from abrasion or drying during handling and while anaesthetised.

Two injectable anaesthetic protocols were identified that would meet maximum injection volume limits (10 ml per intra-muscular (IM) injection site), and that were suitable for abdominal surgery.

One pig received Protocol 1, which consisted of a mix containing 2 mg kg^{-1} body weight each of tiletamine HCl, zolazepam HCl, ketamine HCl, and xylazine HCl (Table 1).

This combination is commonly used for immobilisation and surgery in pig research and fieldwork (Ko et al. 1993; Veterinary Support Personnel Network 2020). Ko et al. (1993) reported that when using this combination, pigs became recumbent within 3 mins, good analgesia was maintained for 24–48 mins, pigs tolerated intubation for 30–50 mins and remained recumbent for 50–70 mins, and then walked with ataxia. The drug mix was prepared as follows: Zoletil™ (Virbac NZ Ltd, Hamilton, NZ) containing 250 mg tiletamine HCl and 250 mg zolazepam HCl as a powder in a vial were mixed with 2.5 ml ketamine HCl 100 mg ml^{-1} solution (Phoenix Pharm Distributors, Auckland, New Zealand) and 2.5 ml xylazine HCl 100 mg ml^{-1} solution (Phoenix Pharm Distributors, Auckland, New Zealand) in the original vial. The dose rate of each active ingredient of 1–2 mg kg^{-1} body weight (0.5–1 ml per 25 kg) via intramuscular injection is recommended, and the higher dose rate of 2 mg kg^{-1} (1 ml per 25 kg) was used in this study. A supplemental dose of 0.5 ml kg^{-1} can be administered if required, or supplemental doses of individual drugs can be administered based on veterinary assessment.

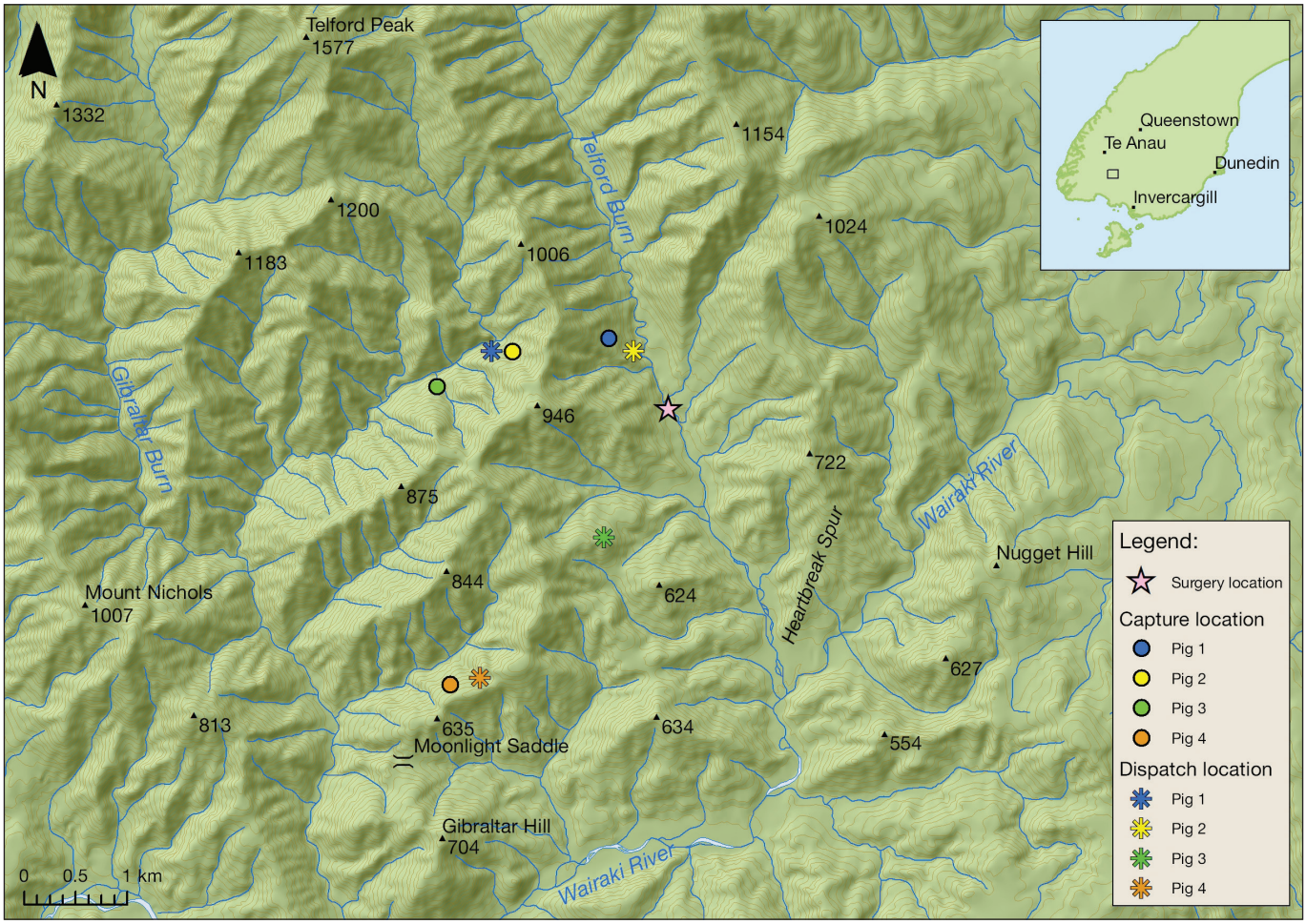

Figure 1. Map of study site. Points show capture and dispatch locations for each individual pig.

Table 1. Drug volumes and concentrations for anaesthesia Protocol 1 combination mix.

Purpose	Drug	Concentration (mg ml^{-1})	Dose (mg kg^{-1})	Volume (ml kg^{-1})
Anaesthetic	Zolazepam HCl	50	1–2	0.02–0.04
	Tiletamine HCl	50	1–2	
	Ketamine HCl	50	1–2	
	Xylazine HCl	50	1–2	
Combination dose rate				
Reversal	Atipamezole*	5 mg ml^{-1}	0.2	0.04
	Flumezenil*	0.1 mg ml^{-1}	0.002	0.02

*As determined by veterinarian

Reversal agents were added to the protocol to shorten recovery time and counteract complications with respiratory depression and hypothermia (Lu et al. 2011) and delivered by slow IV injection. Atipamezole HCl (Antisedan™ Zoetis NZ Ltd, Auckland, New Zealand) at 0.2 mg kg^{-1} was used to reverse xylazine and very low dose flumazenil (Anexate™ Pharmaco NZ Ltd, Auckland, New Zealand) at 0.002 mg kg^{-1} was used to partially antagonise zolazepam. There is currently no reversal available for the dissociative agents ketamine and tiletamine, therefore the anaesthetic is only partially reversed and the side effects of dissociative anaesthesia will still be present during recovery.

Three pigs received Protocol 2, which consisted of a mix of ketamine HCl (10 mg kg^{-1}), medetomidine HCl (0.08 mg kg^{-1}) (Domitor™ Zoetis NZ Ltd, Auckland, New Zealand), and butorphanol tartrate (0.2 mg kg^{-1}) (Butorgesic™ Ilium Ethical Agents Ltd, Auckland, New Zealand) by intramuscular injection (Table 2). This combination can be reversed partially with atipamezole HCl by intramuscular or intravenous injection (Sakaguchi et al. 1996). Sakaguchi et al. (1996) found this combination produced a quick and smooth induction, a duration of effect of 75–90 mins and support of major surgery for at least 30 mins, and the anaesthesia could be quickly and easily reversed with atipamezole. They concluded that this combination is suitable for short-term major surgery in 14–26 kg pigs.

In both protocols, local anaesthetic Nopaine™ (lignocaine 2%, Phoenix Pharm Distributors, Auckland, NZ) was available to locally block pain at surgical incisions or as a 'flush block', whereby it can be irrigated over the surgical area to locally anaesthetise the tissues if required, dosed at 2–4 mg kg^{-1} (Hendrickson et al. 2013). Supplementary doses of butorphanol, ketamine and midazolam (Mylan Midazolam, Mylan™ NZ Ltd, Auckland, New Zealand) were given at the discretion of the veterinarian.

Anaesthetic monitoring

Monitoring data were recorded regularly during the surgery and until recovery commenced and consisted of pulse rate and oxygen saturation (SpO_2) measured with a pulse oximeter on the tongue, heart rate measured by chest auscultation with a stethoscope, body temperature measured with a flexible large-animal rectal thermometer, and respiratory rate, mucus membrane colour, capillary refill time and jaw tone were assessed visually and by palpation. These data were interpreted using normal ranges for pigs both with and without anaesthetic sourced from the University of Minnesota (2021; Table 3).

For recovery, pigs were placed in padded wooden crates and given an IV injection of reversal agent(s) via the ear catheter. When eye movement was observed the IV catheter was removed from the ear. Once recovered sufficiently to hold up their head and stand, the pigs were taken by helicopter in the crate to a flat area near to their original capture site and released.

Supportive care

Intravenous fluids (lactated Ringer's solution or NaCl) were only used briefly to ensure IV catheter patency but were available if needed to manage anaesthetic complications. All pigs were given a broad-spectrum antibiotic (amoxicillin, Betamox® Norbrook Laboratories, Newry, Northern Ireland) at 15 mg kg^{-1} IM and an analgesic (meloxicam, Metacam® Boeringer Ingelheim, Auckland, New Zealand) at 0.4 mg kg^{-1} subcutaneously (SC) towards the end of the surgery.

Surgery

Anaesthetised pigs were placed on a tilted table (see Fig. 2) with hind legs secured to the upper supports and the body placed in either a lateral or supine position. Towels and blankets were used to pad and position the pigs for surgical access and for protection from cold.

The surgical sites were prepared by clipping the hair using battery-operated electric hair clippers then gauze swabs were used to apply chlorhexidine surgical scrub (Microshield 4™, Chlorhexidine 4% Surgical Scrub, Schulke, Macquarie Park, Australia) to cleanse the skin surface, and then sprayed with

Table 2. Drug volumes and concentrations for anaesthesia Protocol 2 combination mix.

Purpose	Drug	Concentration (mg ml^{-1})	Dose (mg kg^{-1})	Volume (ml kg^{-1})
Anaesthetic	Ketamine HCl	100 mg ml^{-1}	10	0.1
	Medetomidine HCl	1 mg ml^{-1}	0.08	0.08
	Butorphanol tartrate	10 mg ml^{-1}	0.2	0.02
Combination dose rate				0.2
Reversal	Atipamezole	5 mg ml^{-1}	0.24	0.048

Table 3. Monitoring parameters reference ranges for pigs (University of Minnesota 2021).

Parameter	Normal ranges without anaesthesia	Normal ranges with anaesthesia
Respiratory rate breaths per minute	20–40	6–20
Heart rate beats per minute	70–1800	60–140
Rectal temperature degrees C	37.7–39.1ºC	>36.6ºC
Oxygen saturation (SpO_2) %	>95	>95
Mucous membranes	Pink, not pale, white, gray, or blue	Pink, not pale, white, gray, or blue

Figure 2. Anaesthetised pig restrained in a supine position on titled table, to allow for saliva drainage and draw the abdominal organs cranially for surgery.

isopropyl alcohol. Sterile disposable surgical drapes were used to protect the surgical site from contamination. The surgeon undertook a hand scrub using a surgical scrubbing brush (BD E-Z Scrub™ Surgical Scrub Brush impregnated with 4% Chlorhexidine Gluconate, BD, USA) and rinsed with fresh water poured from a bucket by an assistant. The surgeon dried their hands using a sterile hand towel and donned sterile surgical gloves for the procedure.

Tubal ligation and resection were preformed using haemoclips (Vesocclude medium/large size ligation clips, Vesoclude Medical LLC, Raleigh, NC, USA) for ligation and surgical cautery to remove a 2 cm section of fallopian tube. The method was chosen for simplicity and speed to minimise surgical time, risk of adverse events and to create non-reversible surgical sterilisation.

A ventral midline approach was used with the incision approximately 7 cm cranial to the pelvic rim and extending 8 cm in length cranially. The ovary was located by hand by palpating adjacent to the pelvic rim, catching the ovary ventromedially. The fallopian tubes were exteriorised and two haemoclips were applied approximately 2 cm apart. The fallopian tubes were clamped with mosquito forceps. A handheld, battery-powered, high-temperature cautery unit (BVI Accu-temp, Beaver-Visitec International Inc, Waltham, MA, USA) with a fine tip was used to cauterise the proximal and distal portion of the fallopian tubes. The uterine horns were manually traced to the other ovary and the procedure repeated. The linea alba was closed with a simple continuous suture pattern using size 0 USP absorbable monofilament suture material (Luxcryl PDO, Lux Sutures, Weiswampach, Luxemburg) and the subcutaneous tissues were closed with a simple continuous suture pattern using size 1 USP absorbable monofilament suture material (Luxcryl PDO, Lux Sutures, Weiswampach, Luxemburg), both using a swaged-on cutting needle. The dermis (skin) was closed using sterile stainless steel staples (Appose™ ULC Skin Stapler 35W Single Use, USL Medical, New Zealand) and tissue glue (Histoacryl™ Blue Tissue Adhesive Tube, USL Medical, New Zealand) applied to the incision line.

A lateral (flank) surgical approach was also considered but not undertaken on a live pig due to logistical constraints on the day. However, the technique was successfully tested on two dead pigs at retrieval (pigs #2 & #3). The pig was placed in left lateral recumbency. A surgical incision was made over the location of the right ovary (Latrach et al. 2007). The right ovary was located by hand then exteriorised thorough the incision, and the left ovary located by following the uterine horn. The fallopian tubes were examined to determine the location for tubal ligation.

Radio transmitters

Each pig was fitted with 3 transmitters consisting of a radio-collar, radio-ear tag and an implantable transmitter (either subcutaneous or intra-abdominal implantation) purchased from either Lotek (Havelock North, New Zealand) or Kiwi Track Ltd (Havelock North, New Zealand) (Fig. 3).

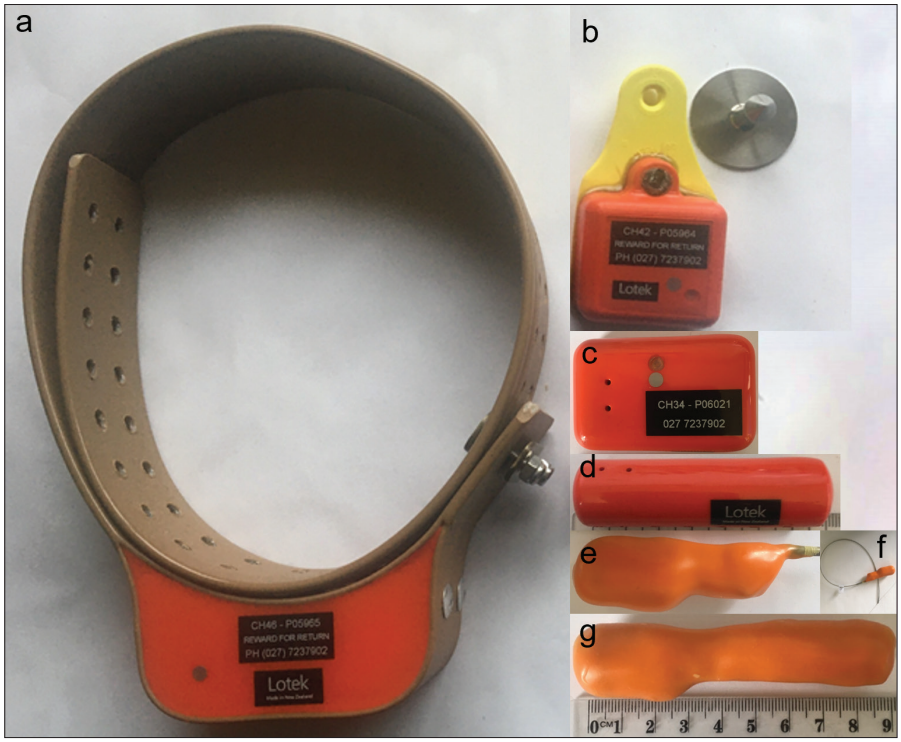

Figure 3. Radio transmitters used in this study consisting of externally attached transmitters (a) collar, (b) ear tag; and implantable transmitters (c) Lotek 154A subcutaneous, (d) Kiwi Track subcutaneous showing body and whipt percutaneous aerial (not to scale), (e) Kiwi Track subcutaneous showing transmitter body, (f) Lotek 163A intra-abdominal and (g) Kiwi Track intra-abdominal.

Table 4. Radio transmitters used in this study.

Manufacture	Type	Model/Product Number
Lotek	Collar	V6C 176D 375-630mm
Lotek	Ear Tag	V2E 154C
Lotek	Implant - Intra-abdominal	V2I 163A
Lotek	Implant - Subcutaneous	V2I 154A
Kiwi Track	Collar	NA
Kiwi Track	Ear Tag	NA
Kiwi Track	Implant - Subcutaneous with percutaneous aerial	350 mm Whip
Kiwi Track	Implant - Intra-abdominal	Internal Loop

Suppliers were requested to programme each transmitter with a different very high frequency (hereafter VHF) radio frequency/channel with transmissions set at 40 pulses per min for normal operation or 60 pulse per min if there was no movement in a 12-hour period (mortality mode) (Table 4). Transmitters had at least 12 months' battery life capacity and were switched on and off with magnets. Transmitters were turned on c. 10 days prior to deployment to identify faults. Implantable transmitters supplied by Kiwi Track were coated in a wax layer to reduce potential adhesions or development of fluid pockets (Horning et al. 2017). Pigs were radio-tracked from a helicopter 10 days after surgery to check transmitter detectability and post-surgical survival.

Prior to implantation, transmitters were sterilized using F10™ SC (F10 Products, Loughborough, UK) veterinary disinfectant at 1:125 concentration for 12 hours, immersed in isopropyl alcohol for 1–4 hours, and then rinsed in sterile saline (sodium chloride (NaCl) 0.9%) just prior to implantation.

Intra-abdominal implantation of two transmitters consisted of simply inserting the transmitter into the abdominal cavity via the surgical incision during the sterilisation surgery.

For subcutaneous implantation of two transmitters, a longitudinal incision of approximately 6 cm was made through the dermis, parallel to and between the dorsal midline and the top of the scapula. The subcutaneous space caudal to this incision was blunt dissected to a length of approximately 8 cm × 3 cm in width, large enough for the body of the transmitter. For the transmitter with an aerial, the technique was based on Mulcahy and Garner (1999), whereby the caudal end of the blunt dissection was then punctured to create an orifice to allow a sterile 10 G French catheter to be passed retrograde into the blunt dissection space. The tip of the catheter was then cut, and the external aerial of the transmitter was threaded through the catheter and the transmitter was inserted into the subcutaneous space with the external aerial protruding through the caudal orifice. The French catheter was then removed. For the transmitter without an aerial, the transmitter was simply inserted into the space created by the blunt dissection. For both transmitter surgeries, the cutaneous tissues were closed using size 0 USP absorbable monofilament suture material

(Luxcryl PDO, Lux Sutures, Weiswampach, Luxemburg) with a swaged-on cutting needle using a continuous subdermal suture pattern. The orifice was closed with a single interrupted suture around the base of the external aerial. Stainless steel staples were inserted over the suture line.

After surgery, all pigs were placed in lateral recumbency on the ground. A transmitter collar was attached by placing the collar around the neck and inserting the locking pin at the belt hole which provided c. 4 cm of space between the collar and the pig's neck to allow for growth but not allow it to pull the collar over the head. The excess collar strap was removed. The ear tag was attached using ear tag pliers and inserted in the midline approximately one-third of the length of the ear on the cartilaginous pinna, avoiding visible blood vessels, and using a stainless-steel backing pin/plate to avoid tag loss.

Dispatch and retrieval

Pigs were aerially tracked with the VHF transmitters and aerially dispatched 104 days after surgery. A competent aerial shooter utilising a semi-automatic .233 accurately placed the shot at an appropriate range to ensure humane and clean dispatch of each pig. Bodies were retrieved to conduct necropsies for determining implantation success and to identify any adverse reactions to the surgery or radio-transmitters.

Results

Capture and restraint

All chase to capture events were of less than 5 mins duration (typically 30 s) and all pigs were caught within 4 km of the surgery site (three pigs were caught west of the surgery site, and one to the south-west; Fig. 1).

Four adult female pigs were captured weighing between 28–43 kg. The pigs lay quietly following capture and did not vocalise or require any additional intervention while awaiting anaesthesia. The restraint technique was very successful in preventing injury to handlers and pigs, and there were no cases of hyperthermia or capture myopathy detected. No tape over the snout was required.

Anaesthesia

No abnormalities were detected during the veterinary external examination of each pig. The IV catheter took between 3 to 5.5 mins to place. In the first pig the catheter became dislodged during handling and was not functional when required for an anaesthetic supplemental dose. In the other three pigs an IV fluid line with sterile saline (NaCl 0.9%) was attached to monitor and promote catheter patency.

Both anaesthetic protocols provided approximately 25 mins of surgical level anaesthesia from the initial induction dose. Anaesthetic supplemental doses were based on the response of the pig and the time remaining for the surgery; thus a variety of combinations were administered across the three pigs which did not adhere to the initial protocol plan and are reported in Table 5.

Protocol 1 was administered to one pig (pig #3). Induction was rapid at <6 mins. A surgical level of anaesthesia was maintained until a tail-flick indicated rising consciousness at 24 mins resulting in a low supplementary dose of anaesthetic being given to briefly extend the anaesthetic time. Both reversal agents were administered. Recovery was prolonged with hypersalivation observed. The pig was alert and sitting at 114 mins post-induction (PI) and fully recovered at release at 179 mins PI.

Protocol 2 was administered to three pigs (pigs #1, 2, and 4). Induction was smooth and rapid at 2 and 4.5 mins in pigs #1 and #2 respectively, and less than 4 mins in pig #4. An anaesthetic supplemental dose was required at 26 min in pig #1 and 27 mins in pig #4. Pig #2 had its surgical procedure completed before 25 mins was reached and did not require a supplemental dose.

Table 5. Summary of anaesthetic results for each individual pig.

Pig ID	1	2	3	4
Capture weight (kg)	40	37.5	43	28
Retrieval weight (kg)	32	38	57	36
Surgery approach	Midline	-	-	Midline
Transmitter implant	Intra-abdominal	Subcutaneous	Subcutaneous (no aerial)	Intra-abdominal
Protocol	2	2	1	2
All times are minutes post-induction of anaesthesia				
Recumbency	2	4 ½	6	<4
Catheter placed	5	10	9	3
Surgery started	17	20	13	20
Suturing closed	38	39	30	50
Supplemental doses	26, 41, 58	-	24	27, 42
On ground	73	40	32	76
Reversal	75	49	37	77
Palpebral reflex	82	49	37	83
Head up	-	52	77	83
Standing	103	100	154	>113

Reversal using slow IV atipamezole at 41–58% of the recommended dose, had a rapid arousing effect with palpebral response (7, 1 and 6 mins respectively), and head movements recorded in pigs #2 and #4 at 3 and 6 mins later, respectively. Standing response varied from 18 to 51 mins after reversal in the two pigs where this was recorded.

Anaesthesia monitoring

There was individual variation between pigs due to differences in surgical position, type of surgery, and anaesthetic supplemental doses. Notably, SpO_2 markedly decreased when pigs were placed on the tilted table and returned to normal when the pigs were laid in lateral recumbency on the ground. The rectal temperature dropped when ambient temperature and wind-chill caused mild hypothermia in pig #4 during the final surgery.

Surgery

Two pigs had the ventral midline approach abdominal surgery for tubal ligation and resection. In both pigs the non-gravid reproductive tract was difficult to locate, standard surgical landmarks were inappropriate for this morphotype of pig, and surgery was only successfully completed in the second pig. A flank approach surgery was not attempted.

Two intra-abdominal transmitters were implanted during the sterilisation surgery in two pigs. Two subcutaneous transmitters, one with a percutaneous aerial, were implanted in the other two pigs.

At retrieval, all pigs were in good condition, with fully healed surgical sites that were indistinguishable from surrounding tissue except for a very slight pucker adjacent to the incision line in the abdominal skin on pig #1.

The flank approach for sterilisation surgery was successfully undertaken on the dispatched pigs #2 and #3 post retrieval. The modified surgical landmarks identified during the field operation were used (Fig. 4).

Retrieval

All pigs were located 104 days after the surgery and release. They were successfully located by tracking the VHF transmitters and humanly dispatched from the helicopter. They were recovered for necropsy.

Transmitter

Operational limitations restricted the testing of all transmitters to a range of approximately 5 km in a straight unobstructed line. At this point all the receiving signals from the transmitters were still strong suggesting the maximum range is significantly further than 5 km, however this range is considered sufficient for the intended application.

The individual transmitter frequencies were selected and submitted to both manufacturers to ensure there was separation between frequencies. Unfortunately, frequencies were mixed and two transmitters from each manufacturer were on the same frequency. Nine out of 12 transmitters were detectable at day 10. Two of the undetected transmitters were not transmitting, which was attributed to the 12-hour duty cycle (a feature to save battery that was not intended for this trial). Eleven of the 12 transmitters were functioning at day 104 when all the Judas pigs were tracked, dispatched and recovered for autopsy. Consistent with the day 10 survey, one ear tag transmitter was not functioning but turned on successfully after it was recovered. Two transmitter collars from one manufacturer were transmitting but were incorrectly in mortality mode (60 pulses per min).

Transmitters were physically investigated after pigs were retrieved. All pigs had their ear tag intact and the surrounding tissue was healthy on visual inspection and palpation. All pigs had the radio-collar intact with a mildly visible indentation in the skin and subcutaneous tissues of the ventral neck. However, all had sufficient space for at least 2 fingers to slip easily under the collar, and a 2–3 mm thick layer of soft mud was present on the ventral collar inner surface on all pigs (Fig. 5).

Both intra-abdominal implants were firmly wrapped in a sheath of 2–3 tissue layers within the ventral edge of the omentum and were situated in the ventral abdomen (Fig. 6). The Kiwi Track transmitter was firmly adhered to slightly thickened omental tissue and could not be separated from the tissue. The Lotek transmitter did not adhere to tissue and once the omentum sheath surrounding it was unrolled, it easily slid out of the tissue. The Kiwi Track subcutaneous transmitter was located during radio-tracking under vegetation c. 1.3 km from the pig's dispatch point. It appeared undamaged and was correctly transmitting in mortality mode. The necropsy of the pig did not reveal any evidence of a wound or infection

Figure 4. Landmarks for flank approach for tubal ligation surgery shown on pig at necropsy. (a) last rib (white arrow) and iliac crest (black arrow), (b) flank incision with uterine horns exteriorised (green arrow), and thumb and forefinger on iliac crest (white arrows).

Figure 5. Images of ear tag and collar on pig #3 at necropsy. (a) stainless steel backing of ear tag, (b) ear tag with transmitter, (c) skin and ear tag puncture, (d) collar in situ with looseness demonstrated, (e) left side of neck, (f) right side of neck.

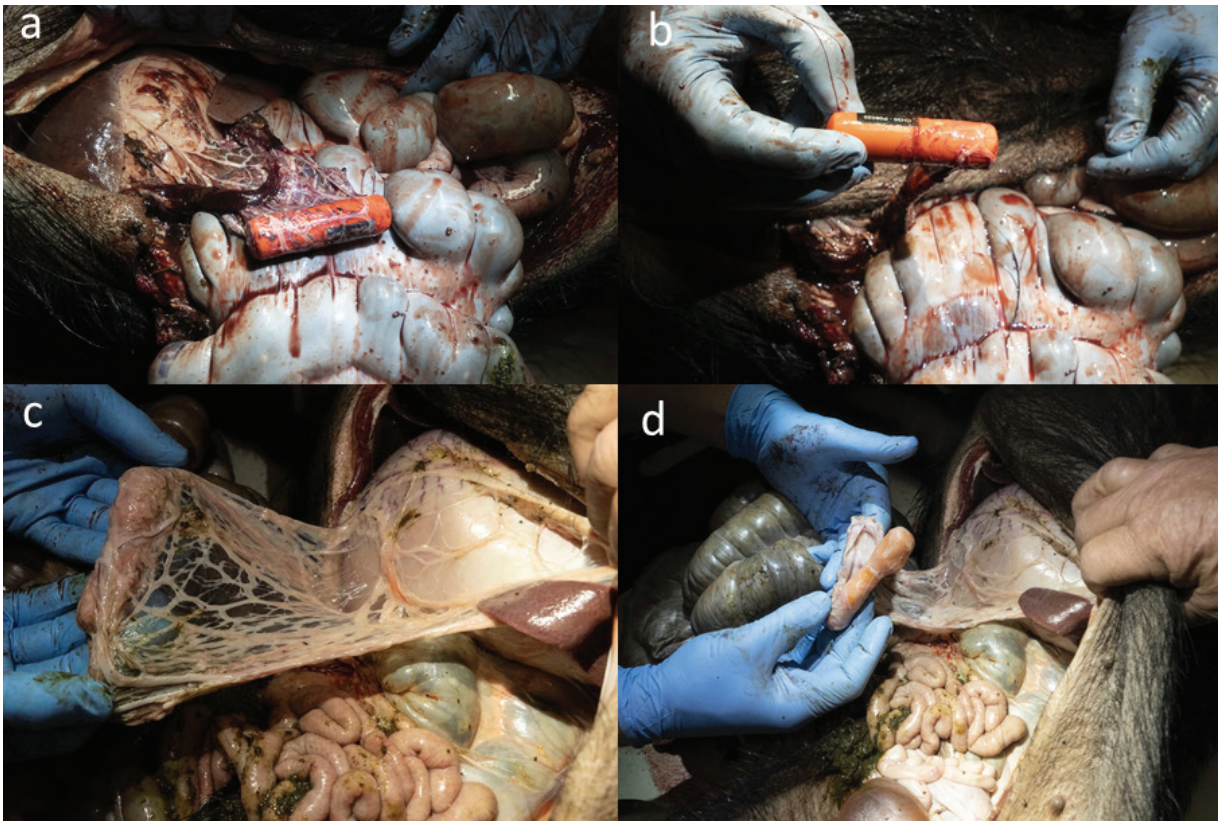

Figure 6. Intra-abdominal transmitters in omentum at necropsy (a) Lotek transmitter in situ, (b) Lotek transmitter partially removed from omentum showing lack of tissue reaction, (c) Kiwi Track transmitter in situ, (d) Kiwi Track transmitter partially removed showing thickened fibrous tissue encapsulation.

associated with the implantation surgery that would explain why the transmitter had fallen out.

The Lotek subcutaneous transmitter was palpable in-situ in the pig where it had been implanted with no evidence of migration under the skin after implantation. It was encapsulated in a non-adherent fibrous layer of tissue within the subcutaneous fat and slid easily out of the capsule when it was incised (Fig. 7).

Discussion

This study revealed some key issues for field application of the techniques used in this study, which can be ameliorated by further refinement of the approach. The capture and restraint methods were effective, however, administration of a sedative at the time of capture would potentially reduce stress to the pigs during the short transport and holding time. This approach was not able to be undertaken during the trial due to legal restrictions on handling of veterinary drugs, and health and safety restrictions on personnel in the helicopter. Alternative capture techniques, such as pen traps, would enable the administration of sedatives at the time of capture and handling, but were not practical for this study.

Open-air surgery in variable weather conditions is not conducive to efficient processing of multiple animals. There were issues with helicopter proximity, sun, wind and cold temperatures, which resulted in disturbance, potential hypo- or hyperthermia, and interfered with sterile surgical processes. A temporary or permanent shelter should be used to reduce these issues in the future.

Each pig had a different transmitter implantation technique, two different surgical approaches were attempted, two different anaesthetic protocols were used, and weather conditions changed from calm and hot to windy and cold as the day progressed. This resulted in the team having to implement different processes for each pig, including the type of recumbency for surgery, location and preparation of surgical sites, duration of anaesthesia, recovery, and body-temperature management. The lack of a repeated routine is reflected in the time taken to commence surgery, which was up to 20 mins in two pigs, the duration of the procedures, and the need for anaesthetic supplemental doses that affected the duration of the recovery times. Once a routine process is established, induction and surgical preparation should be achievable in a much shorter period, which in turn would reduce the need to use subsequent anaesthetic supplemental doses or would clearly establish when such supplemental doses should be pre-emptively administered. Efficiencies will be achieved

Figure 7. Images of Lotek subcutaneously implanted transmitter at necropsy. (a) Palpation of transmitter in situ under skin, (b) incision to show transmitter in subcutaneous fat layer, (c) encapsulated transmitter removed and capsule incised to retrieve transmitter.

when routine application of the same surgical and anaesthetic protocol can be applied to all pigs.

The placement of the IV catheter is a useful tool for fast administration of drugs, but in an otherwise healthy animal going through a routine procedure, it is not necessarily required. The veterinarian can anticipate when IM anaesthetic supplemental doses are required if unable to complete the surgical procedure within the initial period of anaesthesia.

In selecting our anaesthetic protocol, we were aware that there was no ideal drug combination that would deliver all the desirable characteristics in a field setting, namely: intramuscularly injectable, providing a rapid smooth induction, surgical-level anaesthesia for >40 mins duration, and a fully reversible, short, and smooth recovery, that is both practical and affordable. Multiple combinations of anaesthetic agents have been used in studies involving wild pigs for sedation for handling or short surgical procedures (Gabor et al. 1997; Sweitzer et al. 1997; Sutherland-Smith et al. 2004; Barasona et al. 2013; Ellis et al. 2019); however, studies for prolonged surgical-level anaesthesia (>10 mins) tend to be based on domestic or laboratory breeds of pigs for agricultural or experimental purposes (Sakaguchi et al. 1996; Lehmann et al. 2017), or be designed for chemical restraint and induction but not maintenance of anaesthesia (Ko et al. 1993).

We selected two protocols that provided the greatest number of the desirable characteristics, and which used drugs that are readily available in New Zealand. However stronger concentrations of the drugs used are available overseas and could be imported to reduce the volume of the IM injection. The inclusion of dissociative anaesthetic agents was necessary to achieve surgical anaesthesia but is not reversible. This study was not designed to compare protocols, but rather to provide experience with their use in the field and any obvious limitations that need to be considered in designing the eradication programme.

Protocol 1, with a mix of four agents, had a lower dose rate of dissociative drugs per kg body weight with a total of 4.4 mg kg^{-1} of ketamine and tiletamine. Pig #3, which received this protocol and a supplemental dose of ketamine only, had a prolonged recovery with excessive salivation which has been observed with all agents in this drug combination (Muir et al. 2014; Ellis et al. 2019). For this reason, it was only used on one pig, and pig #4 was reallocated to the Protocol 2 group.

Protocol 2 was attractive because of the reported reversibility (Sakaguchi et al. 1996) and suitability for short-term major surgery. Our experience was that this protocol was a predictable anaesthetic, with an approximately 25 min duration of deep anaesthesia suitable for surgical procedures. If surgical time can be reduced to 25 mins, which is feasible for tubal ligation and intra-abdominal transmitter implantation, then this protocol is likely to be sufficient with a single injection. If a longer duration is required, then a pre-emptive anaesthetic supplemental dose of a quarter of the induction dose could be given at 20 mins. Recovery to standing after reversal was approximately 10–20 mins even with supplemental doses of sedatives, which was considerably shorter than Protocol 1 and advantageous in a field situation.

Although the IV route was used for delivery of reversal agents in this study, IM administration of agents would reduce the risk of potential complications from IV administration of atipamezole observed in other species.

Surgical landmarks based on commercial pig breeds were misleading and complicated by unknown age and reproductive status. The first pig was misidentified as gravid and the ventral midline surgical incision placed more cranially to allow for the anticipated anatomical changes to the position of the ovaries resulting in a caudally extended incision line. The fourth and smallest pig (28 kg) was correctly identified as non-gravid with the ventral midline surgical incision correctly aligned with the ovarian position. The flank surgical approach incision is directly over the right ovary, however, locating the ovary was not significantly easier than the midline approach. One advantage is that there is reduced pressure on the wound post-surgery, however, no adverse effects were noted at retrieval in the two pigs that had midline incisions (pigs #1 & #4). If the eradication is implemented, surgical personnel should be provided with culled Auckland Island pigs to practice the sterilisation surgery. This would establish the preferred surgical approach, the correct surgical landmarks for different stages of gestation and ensure surgical proficiency to minimise the anaesthetic time in live animals. Where possible, smaller females should be targeted for easier handling and to improve the efficacy of these procedures.

Monitoring and recording of physiological parameters were undertaken to provide good welfare outcomes for the pigs and understand the impacts of some of the techniques being explored. Time will be saved in a routine situation with multiple pigs being handled in a suitably designed temperature-controlled surgical site, where the use of monitoring methods can be reduced.

The tilt table was considered advantageous to allow for saliva drainage and to draw the abdominal organs cranially away from the surgical site, however, the related drop in SpO_2 was concerning. Because none of the pigs exhibited excessive salivation during anaesthesia, a flat or only slightly tilted table would be more appropriate.

All pigs had a reasonably long recovery time. The pigs were left to recover largely undisturbed, which increased the time to standing recovery. The dropping ambient temperature and increased wind-chill later in the day caused mild hypothermia in pig #4, which likely contributed to its prolonged recovery time. This recovery time is not an issue if the pigs can be held in a protected environment until fully recovered. Excessive salivation, anticipated as an effect of the use of ketamine and Zoletil™ (Muir et al. 2014), was only observed as part of Protocol 1; Protocol 2 is favoured to avoid that side effect.

The results of the necropsy showed that the surgical preparation and transmitter attachment of implantation techniques are suitable for feral pigs with no adverse impacts detected relating to the tubal ligation, ear tag, collar, intra-abdominal or subcutaneous transmitters. The loss of the Kiwi Track subcutaneous implant is puzzling. There was no evidence of an adverse event relating to the surgery or the post-surgical period and the transmitter was undamaged at retrieval. The presence of a percutaneous aerial or the cylindrical shape may have enabled the pig to dislodge the transmitter either accidentally or deliberately but this would require further investigation.

All the working transmitters were found to be detectable at an acceptable level for the purpose of retrieving Judas pigs, which means that transmitter choice can be based on practicalities of product availability, speed of implantation, and security. Intra-abdominal transmitters meet these requirements and a combination of these transmitters with an ear-transmitter is recommended for the Auckland Island eradication. For additional redundancy, a second implantable transmitter could be used if VHF channel separation can be ensured. Careful planning and quality supply are required to ensure unique

transmitter frequencies are employed to avoid confusion during tracking. Having the frequency of the ear-transmitter and intra-abdominal transmitter sequential in one direction on each individual Judas pig will facilitate an efficient transition when checking the transmitters.

The results demonstrate the risk of faulty transmitters. It is critical Judas pigs can be located and controls should be employed to reduce this risk. Transmitters should be turned on at least 2 weeks prior to deployment to check functionality. Consider having a non-latching mortality signal in the ear-tag transmitter only and unless absolutely necessary, duty cycles should be avoided. Transmitters should be meticulously checked before positioning Judas pigs in the field.

Transmitters need to have correct information displayed externally to assist in management and deployment. The collars from the two manufacturers varied in colour with the fluorescent strap significantly easier to recognise. Judas pigs need to be quickly and accurately identified from other Judas or non-tagged pigs so if collars are not used fluorescent tags with letters/numbers in one or both ears (that can be read from 25–30 m) should be used.

This trial highlighted the difficulties faced in a field-based project and provided insight into future design if the programme is applied on Auckland Island. Attention to detail will prove to be critical for the planning and the implementation. Ideally, the surgical staff will have prior experience with pig surgery and an opportunity to develop an efficient routine on culled pigs. Good equipment and surgical site shelter are essential. Although a routine anaesthetic protocol is recommended, having additional drug options available would be useful to allow the veterinarian to adjust the protocol based on any peculiarities encountered when operating on the pigs on Auckland Island. Keeping the transmitters simple with rigorous testing will reduce the risk of transmitter failure. Redundancy with multiple transmitters is recommended with an internal transmitter preventing environmental complications.

Author contributions

FSC, NLM, KM, PH, and MAJ designed the study; FSC, NLM, KM, PH, MAJ, IB and SPL undertook fieldwork; FSC, NLM, KM, PH, MAJ, IB and SPL analysed the data; and KM wrote the manuscript with input from FSC, NLM, PH, MAJ, IB and SPL.

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