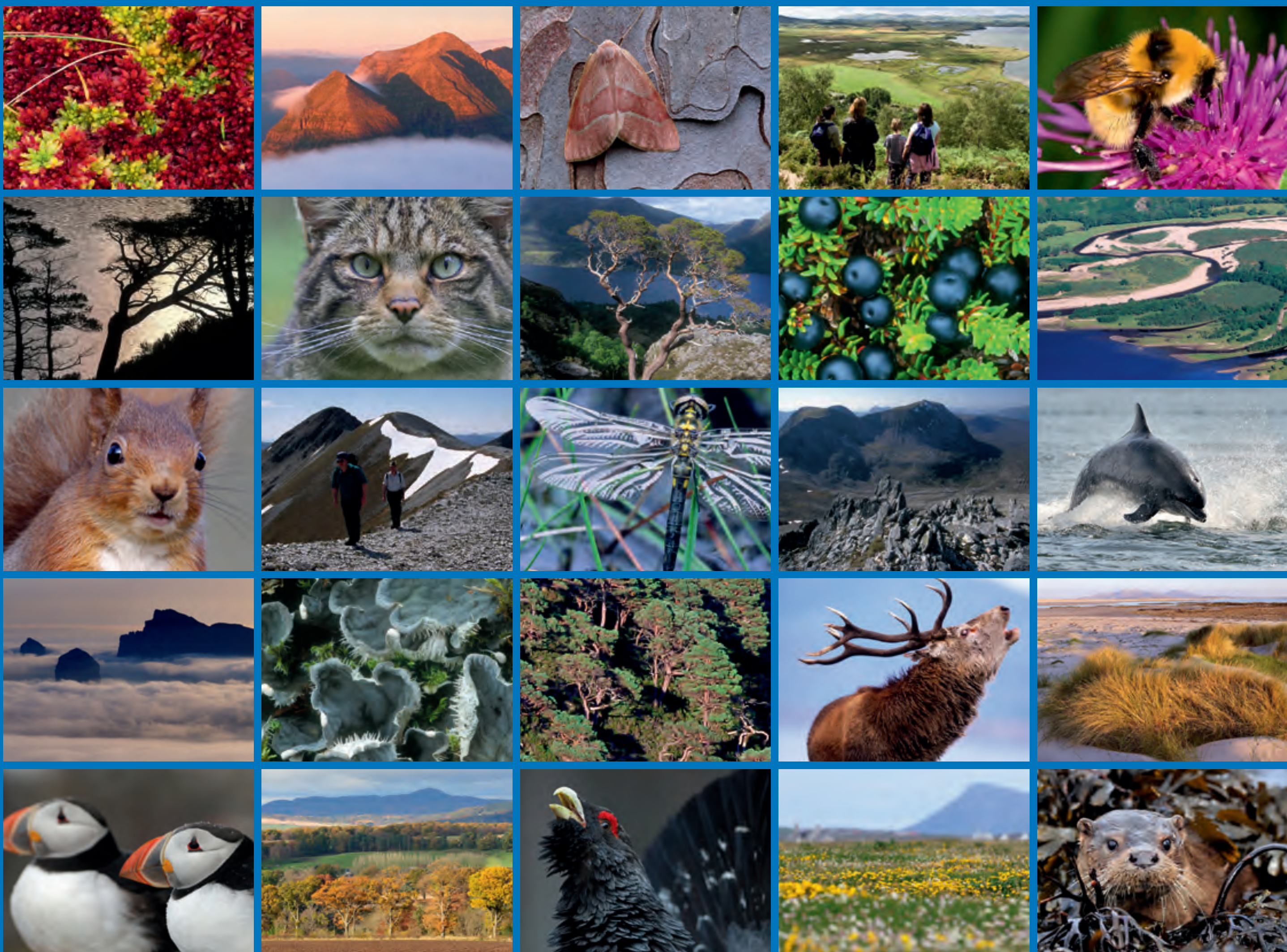
Scottish Natural Heritage
Commissioned Report No. 681

Trapping and health screening of free-living beavers within the River Tay catchment, east Scotland



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COMMISSIONED REPORT

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Trapping and health screening of free-living beavers within the River Tay catchment, east Scotland

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COMMISSIONED REPORT

Summary

Trapping and health screening of free-living beavers within the River Tay catchment, east Scotland

Commissioned Report No. 681
Project No: 13810
Contractor: Royal Zoological Society of Scotland
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Keywords

Beavers; Tayside; Tay; health; trapping; *Echinococcus*.

Background

Since 2006 Scottish Natural Heritage (SNH) has been aware of small scale evidence of free-living beavers in and around the River Tay catchment. A distribution survey undertaken by Campbell *et al*. (2012) determined that a sizeable population was established within this area, outside of the official Scottish Beaver Trial (SBT). In March 2012, the Minister for the Environment and Climate Change announced that the presence of these beavers living in Tayside would be tolerated until a decision on the future of beaver reintroduction to Scotland is made in 2015. Information gathered from SBT, from this Tayside population and from experiences of European counterparts already living with the presence of beavers will be presented to the Scottish Government by SNH to aid in this final decision-making process.

Potentially 38-39 groups of free-living beavers have been identified within the catchment of the River Tay east Scotland (Campbell *et al.,* 2012). The origin and health status of these animals are unknown and may be of concern, particularly if they carry non-native parasites (such as *Echinococcus multilocularis)* or diseases (such as Tularaemia). There is also a need to clarify that the non-native North American species (*Castor canadensis*) has not been released and that any trapped beavers are Eurasian (*C. fiber*). Species identification was recorded here through the colour and viscosity of anal gland secretions, however, further information on species and population identification was undertaken through genetic analysis and will be reported separately (McEwing *et al*., 2015). Reported here are the findings from a beaver trapping and health screening programme which occurred within the Tayside catchment over October 2012 to April 2014.

Main findings

- There is an identified need to assess any disease risk posed by the free-living population of beavers that has become established throughout the River Tay catchment, east Scotland.
- Sampling was undertaken through live-trapping and post-mortem examination. The 21 individuals screened were relatively evenly distributed across the current suspected range.
- The non-native, host specific beaver fluke parasite was present in the majority of individuals (70%).

- No evidence of *Echinococcus multilocularis* or Tularaemia was determined in any live-trapped or post-mortem examined beavers.
- All screened beavers displayed good body conditions, had no physical abnormalities, displayed haematological values within normal ranges and tested negative to all diseases screened.
- Individuals did not display any adverse effects of the live trapping, although minor abrasions to nose, mouth and forepaws were displayed in a minor number of animals on removal from the trap. One beaver had a chipped incisor tooth which may have been caused by biting on the trap. Elevated creatine kinase levels were evident in six individuals, hypothesised to be to be due to increased activity levels from trying to get out of the traps. No individuals displayed any adverse effects during the health screening process itself, although one individual died on recovery from the required general anaesthetic. All other beavers made a full recovery and were released in the late afternoon on the day and at the point of capture.
- One individual was recaptured five months later, ~24km from the previous capture point. This beaver had put on weight and showed no negative effects of previous trapping and health screening procedures.
- From a health and body condition perspective there is no evidence that beavers are failing to cope in a Scottish environment or are suffering from compromised welfare. There is evidence that beavers are subject to mortality from vehicle collisions, and some shooting of individuals is occurring.

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Acknowledgements

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1. INTRODUCTION

The Eurasian beaver (*Castor fiber*) is often described as a conservation success, recovering from near extinction in the late 19th century (Nolet and Rosell, 1998), to a current global population estimated at over one million individual animals (Halley *et al*., 2012). Proactive reintroductions and translocations have been widely used so that this species now occupies much of its former native range, with the exception of Britain, Albania, Greece, Macedonia, Montenegro, Portugal and Turkey (Halley, 2011). Despite this widespread conservation effort relatively little has been published on any associated health screening, disease and mortality considerations. The reintroduction of the Eurasian beaver has been subject to much serious discussion within Britain. Most progress to date has occurred in Scotland which saw the first Government-sanctioned trial reintroduction of this former native mammal in 2009. The Scottish Beaver Trial (SBT) involved the release of five family groups of Eurasian beavers as part of a five-year scientific trial, occurring at a designated release site within the Knapdale Forest, mid-Argyll. All originally-released SBT animals were imported from Norway, underwent statutory quarantine periods and full-health screening measures as described in Goodman *et al.* (2012). The SBT aims to gather information on beaver impacts in a Scottish context, with several independent monitoring partners investigating specific aspects of this. This independent monitoring programme is co-ordinated by Scottish Natural Heritage (SNH) who will report findings to the Scottish Government (SG) in May 2015.

Since 2006 SNH has been aware of small scale evidence of free-living beavers in and around the River Tay catchment, well away from the SBT. A survey undertaken by Campbell *et al*. (2012) determined that a sizeable population was established. In March 2012, the Minister for the Environment and Climate Change announced that the presence of the Tayside beavers would be tolerated until a decision on the future of beaver reintroduction to Scotland is made. Information gathered from SBT, from the Tayside population and from the experiences of other European countries currently living with beavers, will be collated and presented by SNH to SG in May 2015 to aid in the final decision-making process.

There is an identified need to assess any disease risk posed by the free-living population of beavers that has become established across the River Tay catchment, east Scotland, and determine which beaver species is present. No trapping or documented health screening has been previously undertaken, so the origin and health status of these animals is currently unknown. It is suspected that this population is largely composed of beavers that have been born in Scotland to directly imported animals or their descendants, but may potentially include a small component of directly imported animals from Europe that have escaped from private collections. It cannot be excluded that assisted releases may have occurred, over a number of years. A health screening protocol for Eurasian beavers relevant to veterinary considerations concerning Great Britain (as a distinct epidemiological area) has been developed (Goodman *et al*., 2012), and will act as a template for the screening Tayside beavers. This project will also provide samples for genetic analysis to confirm species identification, population origin and genetic diversity, which will be reported separately (McEwing *et al*., 2015).

1.1 Background

Potentially 38-39 groups of free-living beavers have been identified within the catchment of the River Tay, east Scotland (Campbell *et al.,* 2012). The origin and health status of these animals are unknown and may be of concern, particularly if they carry non-native parasites such as *Echinococcus multilocularis, (henceforth abbreviated to EM)* or diseases (such as Tularaemia, *Francisella tularensis*). There is also a need to clarify that the North American species (*Castor canadensis*) has not been released and that any trapped beavers are Eurasian. Species identification was recorded here through the colour and viscosity of anal gland secretions, however, further information on species and population identification was undertaken through genetic analysis and will be reported separately (McEwing *et al*., 2015). Reported here are the findings from a beaver trapping and health screening programme which occurred within the Tayside catchment over October 2012 to April 2014 (this was not the continual trapping period, as it includes site identification and monitoring time).

EM, a taenid tapeworm, has been identified as a particular cause for concern, as it is a pathogenic zoonotic parasite of Central Europe that can prove lethal in humans if untreated (Torgerson and Budke, 2003). As rodents, beavers are potential intermediate hosts, with the normal definitive host for this parasite being the red fox (*Vulpes vulpes*), although domestic dogs (*Canis lupus familiaris*) and domestic cats (*Felis silvestris catus*) may also act as definitive hosts. Concern over the potential of beavers to introduce this parasite to the UK, which is currently deemed as *EM*-free, has risen given the discovery of an *EM* positive beaver in a captive collection in England (Barlow *et al*., 2011). This individual had been directly imported from Bavaria some years previously. There is currently no single ante-mortem 'gold standard' diagnostic test for *EM* in humans, or in other intermediate hosts such as beavers. In human medicine, clinical diagnosis can be challenging, but relies on combinations of imaging modalities such as ultrasound, computed tomography (CT), or magnetic resonance imaging (MRI), serology, nucleic acid detection, as well as surgical examination and histopathology. A combination of several different diagnostic testing methods appears to hold the best potential for the non-lethal screening of beavers.

The risk of wild-trapped beavers introducing *EM* to non-*EM* regions such as the UK depends on the country of importation. For example, beavers released as part of the SBT were imported from Norway where this risk is considered negligible (Davidson *et al*., 2009; Wahlstrom *et al*., 2011), whereas beaver *EM* prevalence in wild Bavarian beavers has been estimated (unvalidated) as ~2.5-5% (Barlow *et al*., 2011), with wild red fox prevalence rates ranging from ~50-80% (Konig *et al*., 2005). There is therefore a risk of beavers imported from *EM* endemic areas carrying this parasite. DEFRA has deemed the likelihood of the establishment of *EM* in British wildlife resulting from beavers imported from *EM* endemic areas as being low but uncertain, due to a number of currently unknown factors, including prevalence rates, husbandry of captive beavers, and the potential of predators scavenging on infected carcasses (DEFRA, 2012). In order to mitigate any future risk of *EM* introduction in association with beaver conservation projects, the most straightforward solution would be to only source animals from *EM*-free countries or use animals that have been captive-bred in Britain.

At the time of trapping and health screening, research into a potential serology test for EM in live beavers was being developed. Samples from all beavers screened in this study (blood from live trapped individuals, tissue/muscle fluid from cadavers), were submitted to the University of Bern for development of this serodiagnostic test. This study was accepted for publication June 2014 (Gottstein *et al*., 2014), and offers a valid alternative for future screening of *EM* in live-trapped beavers.

1.2 Objectives and project timeline

The main objectives of this project were to live-trap a sample of free-living beavers within the Tayside catchment, with the aim of screening 15-30 individuals, which underwent full health screening including ultrasound and laparoscopic investigation for *EM* cysts on internal abdominal organs, particularly the liver, on those animals determined to be at least two years of age and older. This project also collected a range of biological samples for veterinary screening, including parasitology, bacteriology, serology, haematology and blood biochemistry. These samples were collected in order to gather more information on general health status, draw inferences on welfare and adaptability to the environment, and address concerns from the Tayside Beaver Study Group relating to certain notifiable diseases of particular interest to the farming community. A number of specific body measurements were also recorded and body condition described. Samples for genetic screening were also collected, the findings of which will be reported separately (McEwing *et al*., 2015).

Potential trapping sites and landowners began to be identified in October 2012 and health screening protocols were developed, including live-screening for *EM*. From Oct 2012 until April 2014 a continued process of trap site identification, land owner engagement, trap placement, camera trap monitoring and active trapping for health screening occurred.

2. TRAPPING AND ANIMAL HANDLING

2.1 Live-trapping sites

Once a potential trapping site had been identified a site visit was undertaken by Helen Dickinson (TBSG), Neil Mitchell (SNH) and /or Roisin Campbell-Palmer (RZSS). If a suitable area of regular and/or recent beaver activity was established and landowner permission given, then a period of monitoring via camera traps was initiated which often included baiting a site with suitable foodstuffs in order to assess beaver activity and suitability as a trapping site.

2.1.1 Landowner engagement

All landowner engagement was undertaken by SNH, through TBSG, predominantly undertaken and co-ordinated by Helen Dickinson and Neil Mitchell. Potential landowners with beaver activity were identified through a number of approaches including contacts known to members of the TBSG, requests via the TBSG website, articles in the Scottish Farming Leader and local press including the Angus Courier, news articles on the Scottish Land and Estates website, and through completion of a landowner questionnaire aimed at people with beaver activity on their site. Those landowners identified as having beaver activity and happy to engage with TBSG were then visited by Helen Dickinson or Neil Mitchell on behalf of the TBSG in order to confirm recent beaver activity, seek permission to use camera traps to monitor beaver activity and if appropriate, permission to trap and then re-release any beavers back on to the land in question.

Minimal numbers of landowners came forward and securing of potential trapping sites was a slow process. The majority of sites were first identified as having beaver activity from the survey maps produced during the 2012 distribution survey (Campbell *et al*., 2012), and then approached directly by Helen Dickinson. Further sites where identified through suggestions from contacts on the ground and/or various landowners, and these landowners were then approached for the purpose of this study.

In general landowner engagement with this process could be split into;

a) those keen to engage with the trapping process with few reservations.
b) those who were interested in the process but had reservations due to
    i. beaver welfare,
    ii. opinions of neighbouring landowners over their participation in the trap and release process,
    iii. the identification of their land as a beaver site potentially leading to people trying to observe the animals and/or public pressure if they needed to control the population in the future,
c) those reluctant to engage in the process but who would do if their participation was kept anonymous.
d) those who would only engage in the process if any trapped beavers were not returned to their site (these sites were ruled out at this stage).
e) those who did not wish to engage in the process (either didn't want beavers to be trapped at all OR acknowledged they were dealing with beaver management on their own terms).

2.1.2 Trap monitoring

If an area of active and/or recent beaver activity was established and land owner permission granted, then a period of monitoring via camera traps was initiated which often included baiting with food-stuffs. Camera traps were predominantly monitored by Helen Dickinson and Neil Mitchell, with footage analysis by Roisin Campbell-Palmer and Helen Dickinson. Camera trap footage was monitored for a number of variables including level of beaver activity, number of beavers, size and therefore the estimated age class of any beavers present, presence or absence of pregnant or lactating females (determined through the presence of visible nipples), use of the site/trap by any other species (particularly otter), regularity of beaver use, beaver behaviour in and around a non-set trap to flag up any potential welfare concerns (e.g. more than one animal coming to trap at any one time, presence of young or injured beavers, the potential of trap injuries). No species apart from beavers and a single common pheasant (*Phasianus colchicus*) were trapped. There was no evidence of any interest or use by otters (*Lutra lutra*).

2.2 Live-trapping method

All live trapping occurred through the use of *'Bavarian'* beaver traps appropriately placed in areas of beaver activity. All trapping was undertaken under licence granted by SNH, with landowner permission, and involved experienced trappers. Careful consideration of trap placement was taken, especially due to varying water levels and on a number of occasions traps had to be disabled (doors removed so that they could not be set or potentially trap and animals incidentally), due to high water fluctuations. A number of sites had to be ruled out for trapping by this method given topography, difficulty of access with traps, ease of access by members of the public or danger of sudden water level rises.

Figure 1. Bavarian beaver trap placement, trailers and ironhorse were used at some locations to assist trap placement. At other locations traps were carried by hand, involving a minimum of two field staff each time.

Once a trap was in place it was baited with suitable foodstuffs on a regular basis until beavers showed interest and would approach and feed from the trap. This process took from a few days to a number of months, depending on the site. Not all trapping sites had optimal evidence of beaver activity as the number of trapping sites was a limiting factor. In such sites attempts were made to encourage any beavers in the area to use an un-set trap through the use of regular food baiting and on occasion scent from other beavers (castoreum), was trialled to try and stimulate more beaver interest at traps showing little beaver activity.

Figure 2. Bavarian beaver trap set, located near fresh beaver foraging field signs. Note the bait is always placed centrally so that any beaver must enter fully before doors are sprung. In non-set form, doors were removed and food scattered more widely near the water and around the trap to encourage beavers to approach traps.

When regular beaver activity and suitability of animals for trapping were satisfied and veterinary time secured then traps were set. Traps were set in the evening (Figure 2) and checked every morning (7-9am) during their trapping period.

2.3 Transportation of live trapped animals

When a beaver was trapped it was visually observed in the trap to check for any injuries and determine whether it was suitable for transportation. Beavers were removed from the trap with a specifically designed net and placed into a transport crate. All individuals were then transported in a darkened crate (without sedation) with minimal auditory and physical stimulation, to the RZSS veterinary department, Edinburgh Zoo, to undergo a full health screening process.

After health screening procedures were completed each animal was fully recovered from anaesthetic within a transport crate, they were passed suitable for transportation and re-release by the veterinary surgeon. Each beaver was transported in the same manner and re-released at the point of capture. After removal from a trap all animals were transported, examined and returned to release site within ten hours or less. The vast majority of this time comprised travel time from the Tayside area to Edinburgh and back, and recovery time during which the beaver was kept within a darkened travel crate to ensure all effects of anaesthetics had passed, so that the beavers were fit for swimming and able to defend themselves from any potential dangers upon release.

2.4 Cadaver identification and transportation

A general request for notification of any discovered beaver cadaver was made via press pieces in local papers, communications with the Scottish Wild Beaver Group, the TBSG website, and during any landowner approach. A low number of landowners had indicated that they intended to shoot beavers on their land. Alternative management options were suggested but if this course of action was implemented then they were asked to notify SNH and allow cadaver collection for health screening purposes. Five reports of road-killed individuals were made during the trapping period, with one resulting in a beaver cadaver being recovered (the remaining four were confirmed as otters). All cadavers were transported under licence, using appropriate barrier techniques. Post-mortem examinations were undertaken on the day of, or within one-two days of collection, and cadavers were kept in refrigerated stores until the veterinary surgeon was available.

2.5 Trapping effort (from March 2013-April 2014)

Between March 2013 to April 2014, 52 trapping nights were undertaken. During this period traps were set a total of 175 times across 19 sites, with a total of 17 beavers live trapped.

Trapping effort also includes pre-trapping activities:

Pre-trapping site visit

- assessing a site for suitability of trapping (recent beaver activity, site accessibility)
- discussions with landowners to secure permission
- one project staff (occasionally two)
- 37 separate site visits occurred

Trap placement and re-location

- placement of trap at a site
- relocation of trap as required
- minimum of two project staff (on occasions three or four people were involved to assist with difficult placements)
- use of specific vehicles (large enough to transport traps and suitable for off-road use), and equipment such as trailers and iron horse
- 29 separate site visits for this activity took place

Site baiting and camera trap checking

- baiting a site/unset trap with food to encourage and monitor beaver activity
- short duration site visits
- one project staff per site visit on a regular basis
- also supported by various landowners and volunteers who regularly watched beaver activity at various potential sites
- 175 separate checks were made by project staff

Trap setting and checking

- setting a trap and then checking it the following morning
- split between two project staff per day to enable more traps and/or larger distance to be covered
- ~ 52 separate check days were made (between one and ten traps attended per day, across two project staff)

3. HEALTH SCREENING METHODS

3.1 Live animal testing

The disease screening protocol for live beavers was based on that already established for the official Scottish Beaver Trial (Goodman *et al*., 2012), based on guidelines for reintroductions and translocations of wildlife by the Office International des Epizooties (OIE) and World Conservation Union (IUCN) (Woodford, 2001). In addition to this, a number of other diagnostic tests were undertaken to address specific concerns raised by members of the Tayside Beaver Study Group, particularly relating to livestock diseases including Johne's disease (*Mycobacterium avium* ssp *paratuberculosis*) and TB (*Mycobacterium bovis*), following standard laboratory methods. Full and detailed post-mortem examinations were performed on any cadavers of beavers that could be attained, again according to the established protocol (SBT protocols unpublished; Campbell-Palmer and Rosell, 2013).

An initial veterinary visual assessment of the trapped beaver was performed as to the relative safety and suitability of anaesthesia. Anaesthesia was induced by the application of isoflurane in 100% oxygen via facemask while the beaver was restrained. The beaver then underwent endotracheal entubation with a portex uncuffed endotracheal tube under visualisation after local anaesthesia of the glottis with xylocaine spray. Anaesthesia was maintained with isoflurane in oxygen via intermittent positive pressure ventilation, while vital parameters such as heart rate and rhythm, respiratory rate and rhythm, peripheral pulses, and oxygenation were monitored throughout anaesthesia.

A full physical examination was undertaken whilst anesthetised, including:

- eyes – symmetry of head, eyes for ocular discharge
- ears – check for parasites
- nose – check for nasal discharge, abrasions
- teeth – check for malocclusion, signs of dental disease, abdominal wear, trap injuries
- integument (including tail and feet) – check for wounds, ectoparasites, dermatitis, condition of fur, feel for fat cover of pelvic region, spine and tail
- tail – check for wounds, abrasions, thickness

Each beaver was assessed for scars or any signs of previous trauma, as well as for the presence of any external parasites, such as ticks or the beaver beetle *Platypsyllus castoris* (Duff *et al*., 2013). Palpation was performed of the all the limb joints as to normal range of motion, along with an abdominal palpation for any organ enlargements or abnormal masses. Fur condition was assessed as lack of proper grooming may represent underlying health issues and poorer body condition.

Weight was taken on digital scales and body score assessed according to the standard rodent body scoring system adapted for beaver morphology (Campbell-Palmer and Rosell, 2013). Each beaver was scanned for the presence of an identity microchip, and if not present beavers were microchipped in the inter-scapular region to allow future identification (except for four individuals trapped on privately owned land, which were not chipped at the landowners request).

Sex was initially established through the examination of the colour and viscosity of the anal gland secretions (AGS), a recognised field technique given the lack of external genitalia in beavers (Rosell and Sun, 1999). The sex of each individual was further confirmed through abdominal radiographs, abdominal ultrasound, and laparoscopy when performed. Given that beaver AGS also display differences between the two species, it can be used in species identification if the sex of an individual is known (Schulte *et al*., 1995; Rosell and Sun, 1999), so species was identified using AGS as a check that North American beavers were not being released. However, blood samples were taken for genetic analysis to confirm species and reported separately (McEwing *et al*., 2015).

A single channel electrocardiogram (ECG) was performed to assess heart rate and detect any rhythm abnormalities, in conjunction with cardiac auscultation to detect heart murmurs, which are previously reported as a frequent finding in Eurasian beavers under anaesthesia (Devine *et al*., 2012).

Blood was taken aseptically from the ventral tail vein for diagnostic testing and an additional sample taken for genetic screening (reported separately, McEwing *et al*., 2015). Blood parameters and serum chemistry are commonly used in health assessments for a wide range of wild and domestic animals (e.g. Fowler and Miller, 2003; Kaneko *et al*., 2008; Thoresen et al., 2009). Baseline values for wild Eurasian beavers have not yet been published, but have been determined by RZSS (Girling *et al*., unpublished). Published values do exist for other rodents and the North American beaver (Kitts *et al*., 1958; Stevenson *et al*., 1959; Clark and Olfert, 1986; Bennett *et al*., 1991; ISIS, 2002), which will also enable relevant comparisons. Haematology, serum biochemistry, and serum protein electrophoresis were performed as a general assessment of each beaver's general state of health (SAC Consulting Veterinary Services, Scotland's Rural College). Parameters tested are listed in Table 2.

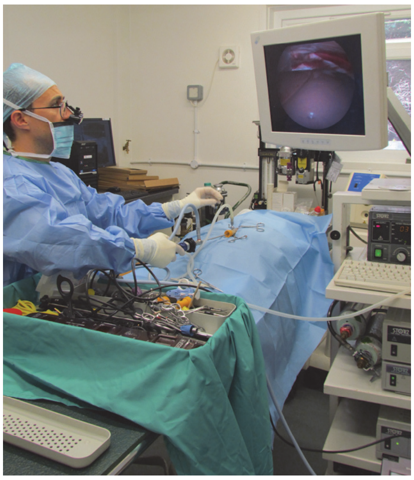
Further specific serological testing was performed. Testing for European *Leptospira* serovars was by means of the microscopic agglutination test (MAT) (Animal Health Veterinary Laboratory Agency, Weybridge). Testing for tularaemia was performed by means of polymerase chain reaction (PCR) (National Veterinary Institute, Norway). Testing for *Echinococcus multilocularis* was performed serologically by means of two different enzyme-linked immunosorbent assays (ELISAs) targeted against the EM 18 and EM 2 antigens, used for human *EM* diagnosis, as well as a recently developed immunoblot. A specific anti-beaver IgG conjugate was used for testing (Gottstein *et al*. 2014), at University of Bern, Switzerland. These *EM* results were interpreted in conjunction with abdominal radiography, ultrasonography and laparoscopic examination findings.

Faeces were taken directly from the beaver's rectum, and rectal microbiology swabs were taken. Faecal samples underwent flotation with saturated salt solution for nematodes and sedimentation for trematodes, including the caecal beaver trematode *Stichorchis subtriquetrus,* previously described in a beaver from the Tayside region (Campbell-Palmer *et al.,* 2013), as well as microscopy for coccidia, *Cryptosporidium* spp., and *Giardia* spp. Standard microbiological culture for bacterial enteric pathogens, including *Salmonella, Campylobacter* and *Yersinia* spp. was performed (SAC Consulting Veterinary Services, Scotland's Rural College). In addition, testing for Johne's disease was requested by the TBSG National Farmers' Union of Scotland representative, and was performed by means of modified acid-fast staining of faecal smears and Polymerase Chain Reaction (PCR) testing (SAC Consulting Veterinary Services, Scotland's Rural College).

Radiographs were taken of the chest and abdomen from two perpendicular views, a dorsoventral and a left lateral recumbency view, as part of the general examination for abnormalities or pathology (Figures 12 & 13). In addition, a bronchoalveolar lavage was performed when possible, as stakeholders had also requested testing for *Mycobacterium bovis*, although the disease has not been reported in beavers. Lavage fluid was submitted for standard microbiological culture and examined cytologically, including acid-fast staining for abnormalities or suspect organisms (Veterinary Pathology, University of Edinburgh).

Abdominal ultrasonography was performed (Figure 14), with specific attention to the liver, to detect any abnormalities that could be indicators of *EM*. A 2-5MHz frequency convex abdominal ultrasound probe was used, and examinations recorded on a digital video recorder. Ultrasonography was performed by wetting the dense fur with 90% ethanol to allow adequate contact and good visualisation, in preference to clipping of fur, which it was considered may adversely affect the beavers waterproofing and thermal insulation when returned to the wild after testing. One limitation of ultrasonography in beavers is that the voluminous gastrointestinal tract frequently contains gas which obstructs the view of parts of the abdomen (Pizzi *et al*., 2012a; b).

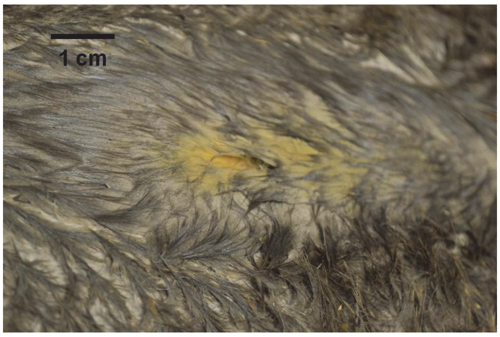
The decision on whether laparoscopic examination occurred was made according to size of an individual, with small beavers still undergoing the majority of the health screening procedures bar laparoscopic investigation for *EM*. In beavers judged to be possibly older than two years of age, a minimally invasive laparoscopic examination of the abdomen (Figure 15) was performed to further assess the liver and abdominal viscera for any signs of *EM* or other pathology not evident on ultrasonography, radiography and physical examination. The laparoscopic technique (Figures 3 & 4) used was that previously successfully used on 18 Eurasian beavers in an enclosed lake, with no post-operative complications recorded when the group was followed for two and a half years (P. Carter personal communication, Pizzi *et al*. 2012a). Laparoscopic examinations were performed by the veterinary surgeon who had performed all previous beaver laparoscopic examinations, and with experience of having performed over 600 veterinary minimally invasive surgical procedures.




Figures 3 & 4. Surgical set-up and laparoscopic examination with abdominal organs visualised on screen.

In the early examinations a one cm square region of fur was clipped over the ventral midline umbilical region, but in later examinations no fur clipping was performed (Figure 5), to avoid loss of waterproofing and thermal insulation in their aquatic environment. The fur and skin in the ventral midline region of the umbilicus was thoroughly cleaned and disinfected with a chlorhexidine or povidone/iodine based surgical scrub, followed by the application of surgical ethanol. A six millimetre skin incision was made and the underlying ventral muscles blunt dissected to allow open access placement of a blunt trocar and 5mm cannula. The abdomen was insufflated with eight-ten mmHg pressure using medical grade carbon dioxide. A five millimetre, 30 degree, 30cm paediatric laparoscope was inserted and the abdomen fully examined, with specific attention to the liver. The animal was repositioned in left and right lateral recumbency to allow movement of the viscera, and visualisation of all organ surfaces. If needed, a 3mm paediatric minimally invasive surgical instrumentation was available for manipulation or sampling of tissues. At the end of the minimally invasive laparoscopic examination the abdomen was deflated and the cannula removed and the muscle and skin closed with absorbable monofilament Poliglecaprone suture material in two layers, the skin closure being performed with a buried absorbable intradermal suture placement. Tissue adhesive was applied to the small skin incision wound to aid in immediate post-operative waterproofing. The resultant wound was approximately twice the size of a microchipping wound.

Minimally invasive surgery holds benefits for wild animals, with small wounds, markedly less pain and faster healing compared with traditional open surgery. Animals can also be returned to their natural environment and social group as soon as they have recovered from anaesthesia, leading to less post-operative stress and minimal disruption to social groups and behaviour. Animals also do not tend to interfere with their small wounds as no skin sutures are required (absorbable sub-cutaneous and intradermal sutures are used), with a lower risk of developing post-operative infections or complications. This type of surgery also holds the benefit that a magnified view of the surgical site is provided, and that internal examinations can be recorded photographically for future reference.



Figures 5. View of incision site on closing after laparoscopic insertion, yellow stain marks incision site stained with iodine and fur wetted to aid ultrasound examination.

Figure 6. Typical underside view of clipped fur area and incision scar following open surgery (for sterilisation in this instance). Note sterilisation operations were not undertaken in this study, photograph taken from another, unrelated project in Germany.

Beavers were administered an injection of long-acting antibiotic (amoxycillin) and an injection of non-steroidal anti-inflammatory pain-killer (meloxicam). Beavers were placed in a straw filled transport box for recovery and the endotracheal tube removed once a swallow reflex returned. Beavers were kept warm in this transport box and monitored until sufficiently recovered to allow safe transport for return to their trapping location for release (Figures 7 & 8).

Figures 7 & 8. Beaver release after health screening, at relevant points of capture.

3.2 Post-mortem examinations

Any reported and successfully collected beaver cadavers within the River Tay region (and one individual recovered from the River Beauly), underwent a post-mortem examination. The same detailed external physical examination was performed as for live beavers, including weight, body condition score, sex determination, dental examination, examination for signs of trauma, injuries, and for presence of external parasites.

The carcass was radiographed for a more detailed determination of skeletal trauma and fractures as well as any signs of being shot. Two perpendicular lateral and dorsoventral views were taken of the chest, abdomen and head. If shooting was determined as cause of death the shot position and type, from which distance and method of dispatch could be determined in order to provide further information for the development of welfare-friendly control recommendations. The locations and details of any landowners coming forward with cadavers were treated confidentially.

The cadaver was then opened and a standard gross post-mortem examination performed with examination of all chest and abdominal organs performed. The gastrointestinal tract was opened and examined for the presence and number of internal parasites. A sample of muscle was stored frozen for genetic analysis.

Faecal samples underwent flotation with saturated salt solution for nematodes and sedimentation for trematodes, including the caecal beaver trematode *Stichorchis subtriquetrus,* as well as microscopy for coccidia, *Cryptosporidium* spp., and *Giardia* spp*.* *S*tandard microbiological culture of faeces was performed for bacterial enteric pathogens, including *Salmonella*, *Campylobacter*, *Shigella* and *Yersinia* species (SAC Consulting Veterinary Services, Scotland's Rural College). In addition, testing for Johne's disease was performed by means of modified acid-fast staining of faecal smears and Polymerase Chain Reaction (PCR) testing (SAC Consulting Veterinary Services, Scotland's Rural College).

Samples of heart, lung, liver, spleen, kidney, stomach intestine, reproductive tract and gonads were taken from any cadaver and preserved in 10% neutral buffered formal saline and underwent detailed histopathological examination by an external specialist veterinary pathologist (Veterinary Pathology, University of Edinburgh).

Blood or tissue samples were tested for tularaemia by means of polymerase chain reaction (PCR) (National Veterinary Institute, Norway). Blood was also tested for *EM* serologically by means of two different enzyme-linked immunosorbent assays (ELISAs) targeted against the EM 18 and EM 2 antigens as well as a recently developed immunoblot, using a specific anti-beaver IgG conjugate (University of Bern, Switzerland). These *EM* results were interpreted in conjunction with any gross and histopathological post-mortem findings as appropriate.

The individual that died on recovery from anaesthetic, following health screening procedures, underwent the full post-mortem examinations described above at an external facility (University of Edinburgh), in order to gain an independent examination to establish cause of death and any pathology present. These findings were then correlated with the live animal examination and testing results.

4. RESULTS

4.1 Physical examinations of live beavers

Seventeen beavers were live-trapped and health screened under anaesthetic. Details of the individual live-trapped beavers examined and the dates of their examinations are given in Table 1.

All live-trapped animals were physically healthy, presented no obvious deformities, external parasites, discharge or obvious signs of disease. Two individuals had evidence of previous, healed wounds (on tail), indicative of beaver-inflicted injuries, most probably as a result of former territorial disputes, which is common and to be expected in this species.

Sex confirmation and examination of AGS both indicated that all beavers examined were Eurasian beavers (Sun and Rosell, 1999), so no individuals were suspected to be the North American species. Therefore full health examination with release was completed for all live-trapped animals.

*Table 1. Live-trapped beavers undergoing health screening. *Beaver 12 was a re-capture, previously identified as beaver 5. This female had dispersed ~24km and gained 2.5kg in the five months since her initial capture.*

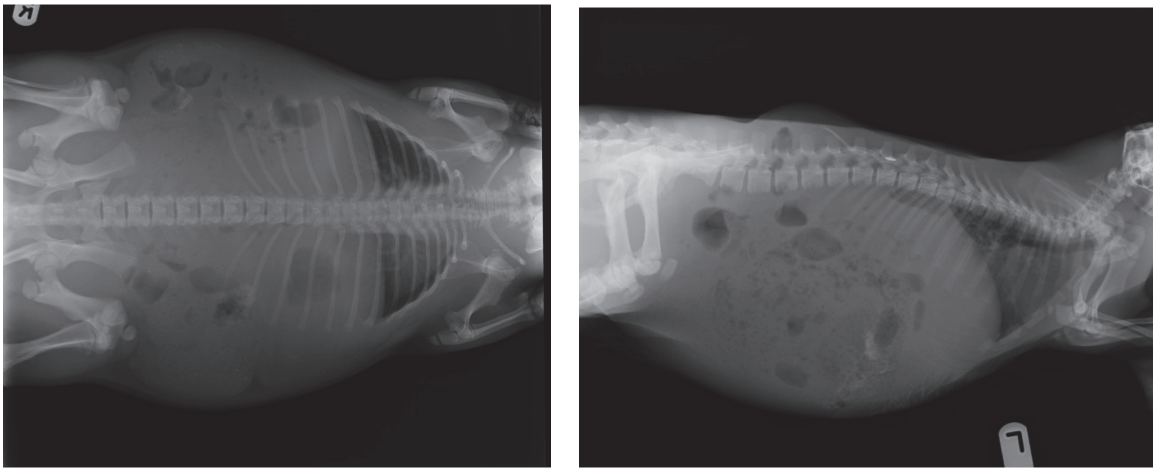
Beaver ID	Live-trapped			
	Trap date	Sex	Weight (kg)	Est. age class
1	06/03/13	Male	10.2	Sub
2	11/03/13	Male	12.8	Sub
3	11/03/13	Male	15.3	Adult
4	27/04/13	Female	12.5	Sub
5	17/06/13	Female	12.5	Sub
6	03/08/13	Male	12.0	Sub
7	04/08/13	Male	22.5	Adult
8	23/09/13	Male	7.7	Kit
9	30/09/13	Female	19.1	Adult
10	09/10/13	Female	7.1	Kit
11	16/10/13	Female	10.2	Sub
12*	27/11/13	Female	15.0	Sub
13	04/12/13	Female	9.0	Kit
14	04/04/14	Male	21.6	Adult
15	26/02/14	Male	13.9	Sub
16	08/03/14	Male	15.8	Adult
17	10/03/14	Female	21.5	Adult

Estimation of age class was made according to time of year, weight and body length. Variation in age classification of beavers according to weight does exist across European populations but weights and age class estimates reported here fell within previously reported values (Pilleri et al., 1985; Hartman, 1992; Parker et al., 2001), though no distinction between yearling and sub-adult was made. Kits were classified as those <10kg within the same year as they were born. Individuals were classified as a sub-adult if <15kgs and/or if female but demonstrating no signs of reproduction (no visible nipples or any indication of previous pregnancies as judged by the ovaries and reproductive tract on laparoscopic examination, and/or evidence that bone growth plates were still open (growing animal) on radiographs. Beavers were classified as adults if >15kgs and/or showing signs of previous reproductive activity and/or fused bone growth plates.

All beavers screened were given body score conditions within the normal range (-3, 3 or 3+), defined as being within normal expected condition for the time of year trapped. This was evident as the vertebrae and dorsal pelvis were not prominent and could be felt with application of slight pressure, tail arches were slightly visible but tails were thick and sturdy, indicating a good, healthy fat layer (Campbell-Palmer and Rosell, 2013).

Two beavers, and adult male and a sub-adult female demonstrated an unusual arrow head shaped incisor wear (Figure 9). A number of the other adult beavers had increased wear to one side of the upper incisors (Figure 10). All beavers had completely normal back teeth and wear patterns. This pattern of wear is unusual in rodents, and likely related to their gnawing and feeding behaviour over a long period of time, and unrelated to trapping. It is unclear what natural history factors may have contributed to this wear pattern, but there appeared to be no related adverse effects.

Figures 9, 10 & 11. Unusual tooth wear found, figure 9 (left) shows 'arrow head' shaped incisor wear, figure 10 (centre) demonstrates increased tooth wear to one side, and figure 11 shows incisor damage (right). Given the mis-shape and whiteness of the inner tooth it is presumed this was caused whilst the beaver was within the trap, most likely by biting a metal surface within the trap in an escape attempt. Tooth root was not exposed so this injury was not determined to be causing pain or interfering with feeding, so the beaver was released following the health check. Incisor re-growth in beavers is quite rapid, so this chip is presumed to be unnoticeable in a matter of weeks (this is supported by previous experiences with captive beavers, RZSS pers. comm).



Figures 12 & 13. Diagnostic imaging consisting of chest and abdominal radiography.

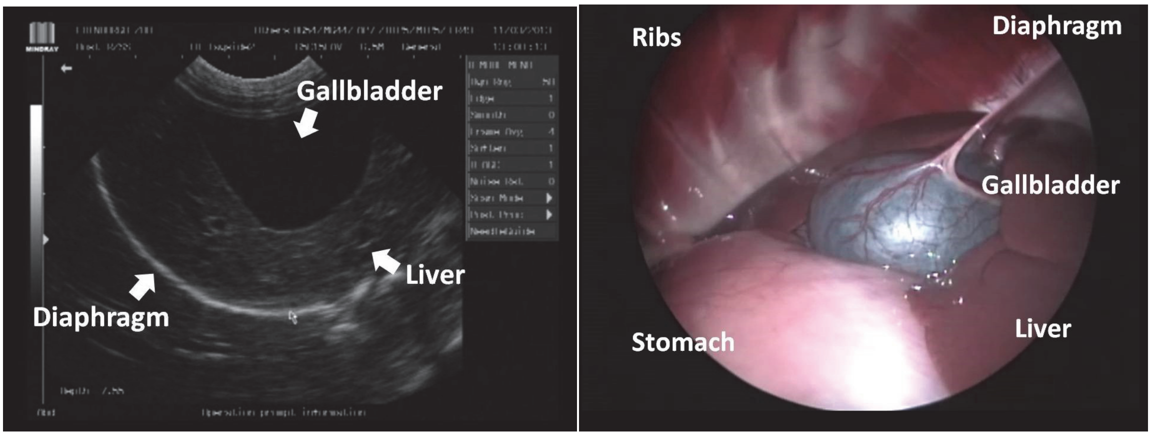


Figure 14 & 15. Normal dorsoventral and lateral chest and abdominal radiographs, abdominal ultrasound and minimally invasive laparoscopic examination view of abdominal organs.

All abdominal ultrasound and laparoscopic examinations were normal with no indications of *EM* infection.

4.2 Haematology, serum biochemistry and plasma protein electrophoresis

Parameters were judged against previously established normal values for the Eurasian beaver (Girling *et al*., unpublished), as well as against the normal reference ranges for North American beavers (Kitts *et al*., 1958; Stevenson *et al*., 1959; Clark and Olfert, 1986; Bennett *et al*., 1991; ISIS, 2002). Haematology, serum biochemistry, and protein electrophoresis parameters tested are listed in table 2. All parameters were largely unremarkable in all live beavers tested, with the exception of six beavers that had elevated creatinine kinase. In the absence of any other abnormalities in other blood parameters or on physical examination, it appears likely these were a result simply of muscle activity of trapped beavers as they attempted to get out of traps. The normal references values used for comparison are likely to be lower than seen in these tested beavers, as they consist of values of long term zoo animals or beavers caught by different methods.

No haemoparasites were recorded in this study, this reflects previous work undertaken on wild Norwegian beavers (Cross *et al*., 2012).

Table 2. Haematology, serum biochemistry and serum protein electrophoresis parameters screened.

Haematology parameters measured	**Serum biochemistry parameters measured**
Red Blood Cell count (x 10^{12}/L)	ALT (iu/L)
Haemoglobin (g/L)	ALP (iu/L)
Packed Cell Volume (PCV) (L/L)	AST (iu/L)
MCV (fL)	Gamma GT (iu/L)
MCHC (g/L)	Bile Acids umol/L
Platelets (x 10^9/L)	Bilirubin (total) umol/l
White blood cell count (x 10^9/L)	Creatinine Kinase (iu/L)
Neutrophil-segmented (x 10^9/L)	Urea (mmol/L)
Band form neutrophils (x 10^9/L)	Creatine (umol/L)
Lymphocytes (x 10^9/L)	Total protein (g/L)
Monocytes (x 10^9/L)	Albumin (g/L)
Eosinophils (x 10^9/L)	Globulins (g/l)
Basophil (x 10^9/L)	Cholesterol (mmol/L)
Blood smear examination	Amylase (iu/L)
	Lipase (iu/L)
Serum protein electrophoresis measured	Calcium (mmol/L)
	Phosphate (mmol/L)
Albumin (g/l)	
Alpha-1 globulin (g/l)	Potassium (mmol/L)
Alpha-2 globulin (g/l)	Sodium (mmol/L)
Beta-1 globulin (g/l)	Chloride (mmol/L)
Beta-2 globulin (g/l)	GDH (iu/L)
Gamma globulin (g/l)	Magnesium (mmol/L)
	BOHB (mmol/L)

4.3 Post-mortem examinations

A single live trapped adult female beaver (ID No 9) died on recovery from anaesthesia. The beaver was submitted for a full and independent post-mortem examination and histological examination at the Department of Veterinary Pathology, at the Royal (Dick) School of Veterinary Studies, the University of Edinburgh. All diagnostic testing of this animal while alive was unremarkable, as was external blood testing at Scotland's Rural College. Unfortunately the cause of death could not be determined from the detailed post-mortem examination and testing, with only mild abnormalities evident. There were some multifocal acute petechial haemorrhages in the grey matter of the brain in the thalamus and brainstem regions, and some mild lung intra-alveolar oedema. Both of these mild findings are likely to be agonal artefactual changes. There was no sign of any problem related to the beaver's capture or health screening procedure. A further detailed review by the RZSS veterinary department could also not find any explanation for the loss of this beaver. Anaesthesia always carries a risk, whether in people or wild animals. It appears this was one of those rare cases with no recognisable cause.

Six beaver cadavers were submitted for post-mortem examination. Two immature males in good body condition were found to have died from severe trauma consistent with motor vehicle trauma (Figures 16-18). Four beavers, also all in good body condition, two adult males, an adult female, and an immature male were submitted after being shot (Figures 19-21). The adult females and one male were submitted already skinned and with the head and tail removed, and had been frozen before submission. The adult female, submitted for post-mortem examination in February was early pregnant with three foetuses (Figure 22). Aside from findings of trauma due to motor vehicle collision or being shot, there were no signs of any pathology, notable infectious diseases, or other signs of ill health, and all beavers were in good body condition. Four beavers were found to carry the beaver caecal trematode *Stichorchis subtriquetrus,* which had previously been reported in the Scottish Beaver Trial animals (Goodman *et al*., 2012), and previously in a free-born Tayside individual (Campbell-Palmer *et al*., 2013). The exception was the immature shot male which was clear of the trematode. One shot male beaver was also found to be positive for *Cryptosporidium* on faecal examination. An examination of 910 wild beavers from Norway found all animals negative for this intestinal parasite (Rossell *et al*., 2001), and concluded beavers were not a source of *Cryptosporidium* spp. for domestic animals or humans. It is however possible that a beaver may incidentally be infected from a domestic animal source. As this was the only occurrence of *Crytosporidium* it may be assumed that this is not a 'normal' finding in beavers, whereas this is incredibly common in domestic young cattle, which were present in the near vicinity of the trapping site. There were no signs of the parasite causing any ill health and the individual was in good body condition.

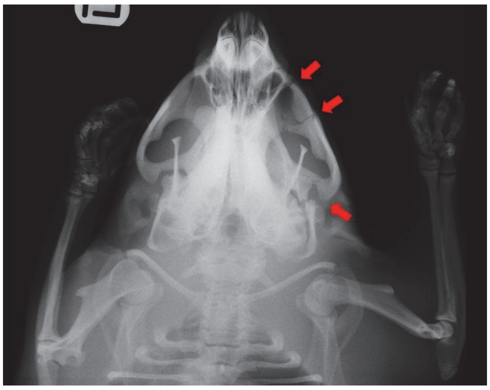
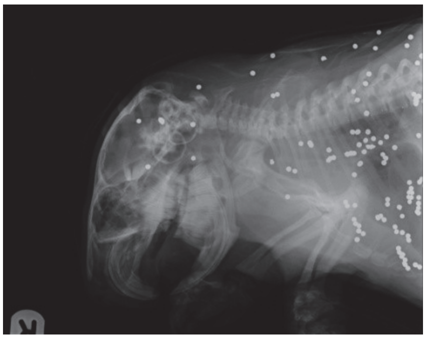


Figure 16 (left), 17 (centre) & 18 (right). Post-mortem radiographs demonstrating skull and limb fractures in two male beavers killed by motor vehicle collisions.



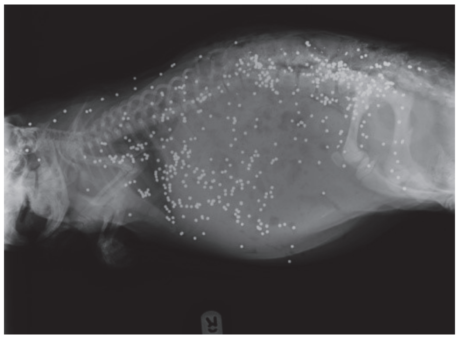


Figure 19 (left), 20 (centre) & 21 (right). Post-mortem radiographs of a shot beaver, demonstrating steel shot at close range.

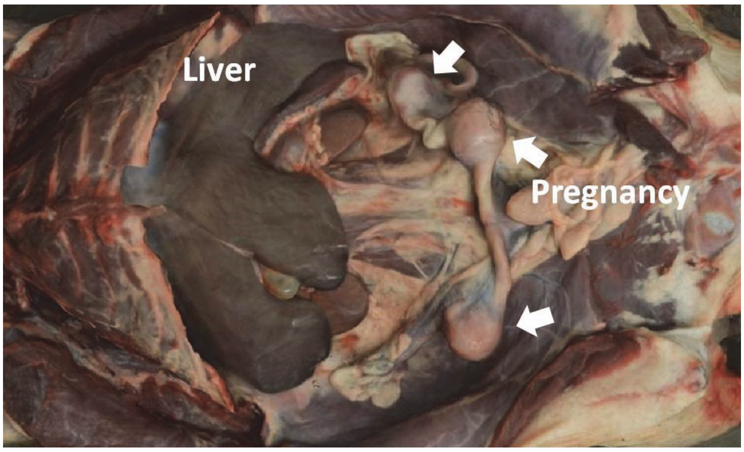


Figure 22. Post-mortem examination of shot adult female beaver demonstrating early triple pregnancy.

At the time of writing beavers may be legally dispatched by landowners as long as they abide by firearms and animal welfare legislation.

4.4 Specific disease testing

No beavers tested, either live or post-mortem examined cadavers, tested positive for any of the following specified diseases: *Echinococcus multilocularis*, Tularaemia, Leptospirosis (Serovar pools 1-6), *Mycobacterium bovis*, *Mycobacterium avium* ssp. *paratuberculosis* ("Johne's disease"), Salmonellosis, or other faecal bacterial pathogens.

Eleven of the live tested beavers and four of the beaver carcasses examined post-mortem tested positive for the beaver caecal trematode *Stichorchis subtriquetrus.* This parasite is the most frequently found parasite reported in beavers and a strictly specialised parasite of beavers (Koubkova *et al*., 2002; Drozdz *et al*., 2004; Vengust *et al*., 2009). Prevalence of this parasite has been recorded as more than 90–100 per cent in beavers (Ahlen, 2001; Drozdz *et al*., 2004). Pathological infection rates tend only to be recorded under high population densities (Borisov, 1941). This parasite has been previously been reported in a young, likely Scottish-born, female beaver from the River Tay catchment (Campbell-Palmer *et al*., 2013), and its eggs have been found in routine faecal screening for beavers released as part of the official Scottish Beaver Trial, mid-Argyll (Goodman *et al*., 2012). It is not perceived as a threat to other wildlife or humans (Koubkova *et al*., 2002). Intermediate host species have been demonstrated to be fully functional when infected (Orlov, 1948; Orlov and Dinesman, 1948). Interestingly, live-trapped beaver 5 was found to be positive when first tested, but when recaptured (as live-trapped beaver 12), tested negative. The infection rate detected in the River Tay catchment in this study of 70% (n= 14 of 20, 95% confidence interval of 48-86%) is likely an underestimate, due to intermittent shedding of the intestinal flukes eggs in faeces tested.

Figure 23. Anaesthetic recovery monitoring, beavers were not released until full recovery was determined, particularly to address potential drowning risk on release.

4.5 Post-health check and release observations

No specific monitoring of individuals following release was undertaken, however, incidental evidence was gathered from three sites where health-screened beavers were observed and individually identified via camera trap footage, direct animal observation and accidental re-trap.

A single live-trapped beaver (ID No 12) was found to be microchipped on scanning, and to have been previously caught and health screened five months earlier (ID No 5). This young female had travelled a distance of ~24km and gained 2.5Kg since first being caught. A physical examination revealed trapping and previous testing did not appear to have had any adverse effects on this beaver, this individual was not re-examined laparoscopically, but radiographs and blood screening were re-run.

Two individuals were ear tagged prior to release, one adult male and one sub-adult male at two separate locations, at the request of the landowner. Both individuals were repeatedly seen on camera traps left in the locations, and through actual observations by people regularly watching each territory. Both were observed regularly (the adult male remains ear tagged and is still seen regularly at the time of publishing of this report). The behaviours of both have remained normal, including feeding, grooming and various social interactions. The adult male is known to have bred both seasons since his capture and health screening. Locomotion in both appears normal (both swimming and walking), with no indication of any pain or discomfort.

5. CONCLUSIONS

There have been no notable findings of ill health, or any infectious diseases of importance to humans, domestic animals or other native wildlife species in the 21 free-living Eurasian beavers from the River Tay catchment examined and tested until the end of May 2014. All beavers examined were in good body condition, with no indications of any being unable to cope with their current environment, or suffering from compromised welfare.

There is evidence of beaver shooting by some landowners within the River Tay catchment. This does raise welfare concerns, and we suggest that prior to the Scottish Government decision on the future of beaver reintroduction to Scotland, advisory recommendations regarding appropriate methods (including calibre and ammunition), and seasonal timing of dispatch should be made to ensure any such deaths are quick, humane and do not result in dependent offspring being left in the lodge.

5.1 Trapping limitations

This study had a number of limitations, largely concerning the trapping aspects, although access to veterinary time should also be considered, as having a veterinary team on permanent standby for extended trapping periods can be resource intensive and expensive.

Bavarian beaver traps are cumbersome in the field and awkward to transport, requiring suitable vehicles and personnel to move them. This can limit the opportunity to relocate traps at short notice and/or in immediate response to beaver activity, it can also limit access to potential trapping sites. Landowner buy-in to the process was slow and ultimately only 27 sites were offered. Some of these were deemed unsuitable for a variety of reasons including lack of recent and regular beaver activity; unsuitable topography (e.g. river banks too steep); accessibility of site and risk from public interference with traps; physical accessibility for trap placement, monitoring and animal removal; and/or hydrological considerations, particularly the risk of trap flooding. There were also occasions where potential trapping sites were identified but landownership was either unknown or permission to trap was not given.

Site monitoring and trap placement therefore occurred on an ‘as and when’ basis, as supposed to the proposed ideal that twelve traps would be in operation at any one time in targeted areas. As a result, it was not until nearer the conclusion of the trapping study that all twelve traps were deployed at the same time on offered sites which were widely distributed. As a result distances between trapping sites were quite large, often requiring more than one field staff to check them each morning. In order to accommodate staff availability, this meant that on occasions some traps in more remote locations where not set as frequently as they may have been. However, the aim of sampling 15-30 individuals was achieved with a good distribution throughout the River Tay catchment. Any future trapping should take into consideration the proportionally higher numbers of trapped and released sub-adult individuals as opposed to adult animals.

5.2 Future considerations for *Echinococcus multilocularis*

EM remains an important consideration when evaluating the health of any beavers reintroduced to Scotland, including those present within the River Tay catchment. It is a zoonotic parasite of serious health concern in Europe, and is regarded as one of the most pathogenic zoonoseis in temperate and arctic regions of the northern hemisphere (Gottstein *et al*., 2001; WHO, 2001). Human infection is usually fatal if untreated (Torgerson and Craig, 2009). Red foxes are the usual definitive host in Europe, but dogs are also highly susceptible to infection (Saeed *et al*., 2006). An extremely high prevalence rate of over 40% in foxes in some areas of central Europe has been reported (Eckert, 1997). While it is established in central Europe, the UK is currently regarded as free from this parasite.

The probability of beavers causing the establishment of *EM* in the UK is currently regarded as a low risk (Kosmider *et al*., 2013), in comparison with the risk posed by the movement of pet dogs from affected regions of Europe (Torgerson and Craig, 2009). A risk assessment found a 98% likelihood of at least one in 10,000 dogs returning to the UK from a short trip to Germany being infected with the parasite if untreated, with over 99% risk if dogs had been longer-term residents (Torgerson and Craig, 2009), hence the requirement for tapeworm treatment of dogs entering the UK (Pet Passport Scheme). There is however a single post-mortem report of an *EM* infected beaver from England (Barlow *et al*., 2011). As the beaver is an intermediate host it should be noted that it must be predated or scavenged after death for an infected beaver to pass the parasite on, whereas the definitive host such as foxes and domestic dogs can pass the parasite on via their faeces.

Whilst there is an anecdotal report of a prevalence rate of 2.5-5% *EM* infection in beavers residing in Bavaria, based on hunters recall from ~400 culled beavers (Barlow *et al*., 2011), the actual prevalence of *EM* in beavers in endemic regions is unknown. The low prevalence of disease considerably increases the risks of misdiagnosis with any single test, and its implications. As an example, assuming a prevalence of five percent, using a single diagnostic test with an 80% sensitivity and 80% specificity, would result in almost one in four tested beavers testing positive (although only one in twenty or five percent are actually truly infected). If this resulted in animals that tested positive being culled, only one in every five culled animals would actually have the disease. This has been demonstrated in a recent study in sheep (*Ovis aries*) in which ultrasound examination for cystic echinococcosis (caused by *E. granulosus*), gave a test sensitivity of ~89% and a specificity of 76% (Dore *et al*., 2014). This complicates both the accurate detection of a low prevalence disease, as well as error rate in its diagnosis and the resultant consequences. The approach comprising a combination of different diagnostic tests used in assessing the Tayside beavers appears to have resulted in an overall sensitivity and specificity approaching 100% in this and other testing (Gottstein *et al*., in press, for example).

Whilst a qualitative risk assessment has been performed on the risk posed by beavers introducing *EM* to the United Kingdom (DEFRA, 2012), it is still currently not possible to quantify accurately the actual risk without further studies. It is recommended not only that the cadavers of any beavers found dead are examined for the presence of the parasite, but also that faeces of the final hosts, red foxes, are collected and tested for the presence of the parasite in the River Tay catchment.

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ANNEX 1: MAP OF TRAPPING AND SAMPLING LOCATIONS (EXCL. DOLLAR AND RIVER BEAULY CADAVERS)

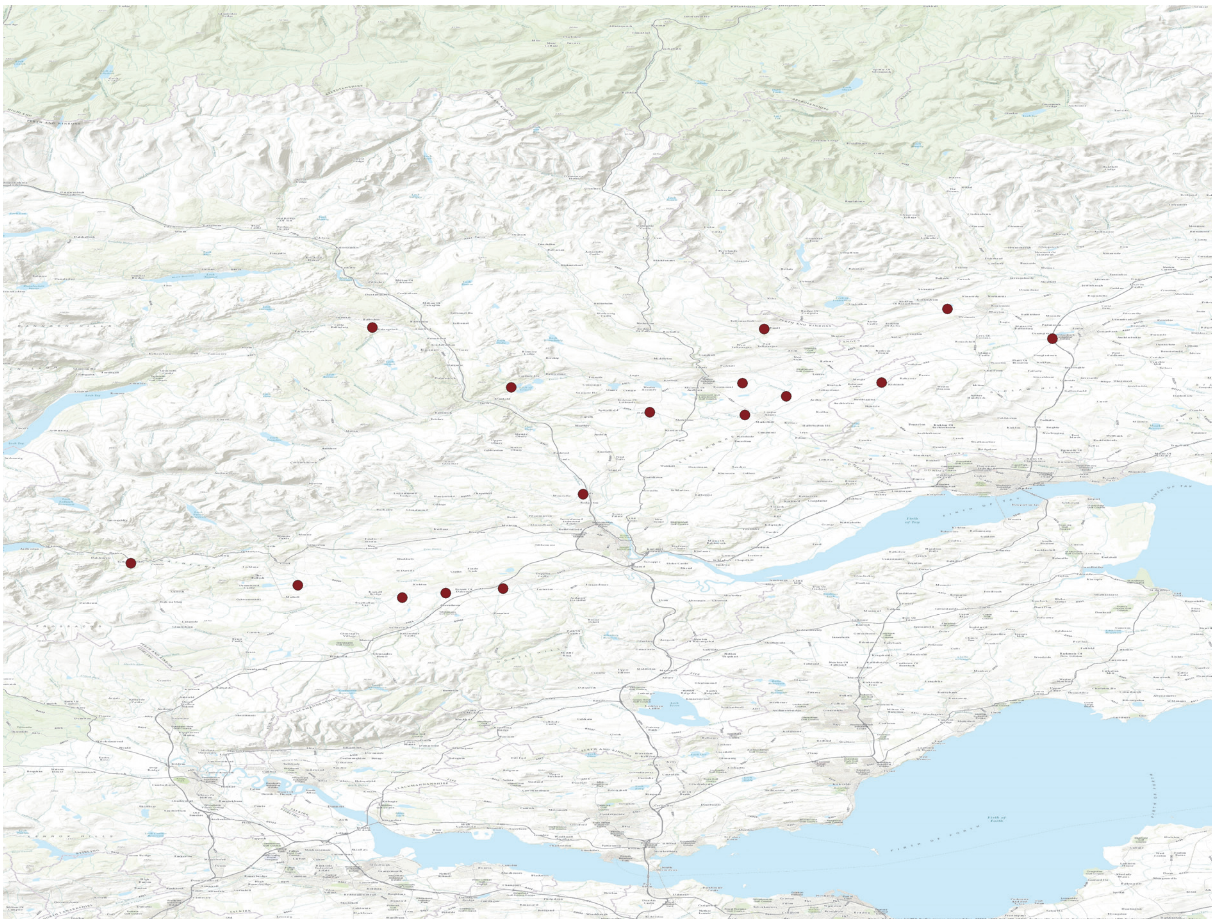

Figure 24. Map of trapping and sampling locations (excl. Dollar and River Beauly Cadavers)

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