



Mayhew, Mic, Chantrey, Julian and Morphet, Nick (2017) Disease and welfare risk assessments for the reintroduction of Eurasian lynx (Lynx lynx) from Sweden to Kielder Forest, Northumberland, UK. (Unpublished)

Downloaded from: http://insight.cumbria.ac.uk/id/eprint/3528/

Usage of any items from the University of Cumbria's institutional repository 'Insight' must conform to the following fair usage guidelines.

Any item and its associated metadata held in the University of Cumbria's institutional repository Insight (unless stated otherwise on the metadata record) may be copied, displayed or performed, and stored in line with the JISC fair dealing guidelines (available here) for educational and not-for-profit activities

provided that

- the authors, title and full bibliographic details of the item are cited clearly when any part of the work is referred to verbally or in the written form
 - a hyperlink/URL to the original Insight record of that item is included in any citations of the work
- the content is not changed in any way
- all files required for usage of the item are kept together with the main item file.

You may not

- sell any part of an item
- refer to any part of an item without citation
- amend any item or contextualise it in a way that will impugn the creator's reputation
- remove or alter the copyright statement on an item.

The full policy can be found here.

Alternatively contact the University of Cumbria Repository Editor by emailing insight@cumbria.ac.uk.

1

Disease and Welfare Risk Assessments for the reintroduction of Eurasian lynx (*Lynx lynx*) from Sweden to Kielder Forest, Northumberland, UK.²
[21]/[03]/2017]

¹ AE Comment – Can we use LUT logo?

AE Comment – Consider renaming to account for welfare and other veterinary issues.

Dr Mic Mayhew BVM&S MRCVS MSc³ Lecturer BSc (Hons) Zoology University of Cumbria Fusehill Street Campus Carlisle CA1 2HH

Dr Julian Chantrey BSc BVMS PhD FHEA FRCPath DipECZM MRCVS Senior Lecturer in Veterinary Pathology University of Liverpool Veterinary Institute Leahurst Campus, Neston CH64 7TE

Dr Nick Morphet BVSc BSc (Hons) MRCVS Veterinary Surgeon 49 Meadowfield Road Stocksfield Northumberland NE43 7PZ

⁻

AE Comment – Move to page 2, see Business Plan.

Contents⁴

- 1 Introduction
 - 1.1 The Lynx UK Trust CIC
 - 1.2 The Proposed Project
 - 1.3 Disease Risk Assessment
 - 1.4 IUCN Guidelines for Wildlife Disease Risk Analysis
- 2 Methodology
- 3 Demographic Parameters of Donor Population
- 4 Translocation Pathway
 - 4.1 Donor and release sites
 - 4.2 Capture
 - 4.3 Transport
 - 4.4 Soft release
- 5 Hazard Categories
- 6 Hazard Identification
- 7 Risk Assessment
 - 7.1 Release, exposure and consequence assessments
- 8 Mitigation measures
 - 8.1 Pre-export
 - 8.2 Transport
 - 8.3 Release pens
 - 8.4 Post release monitoring and intervention
 - 8.5 Post-mortem examination
- 9 Consultations
- 10 Discussion
- 11 Acknowledgements
- 12 References
- 13 Appendices

Appendix 1. Individual Disease Risk Assessments: Infections

- A. Rabies virus
- B. Tick-borne encephalitis virus
- C. Echinococcus multilocularis
- D. Sarcoptes scabei
- E. Francisella tularensis
- F. Generic ticks

Appendix 2. Individual Disease Risk Assessments:Welfare Hazards

- A. Illegal persecution
- B. Generic stress

Appendix 3. Lynx necropsy protocol

Appendix 4. Hazard Identification Spreadsheet

AE Comment – to later be formatted to LUT style.

List of Figures

Figure 1. Steps in the disease risk analysis (IUCN, 2014).

Figure 2. Corine land cover map indicating red boundary of proposed Lynx release area in Kielder Forest.

List of Tables

Table 1. Relative benefits of Kielder Forest as a release site for the trial Lynx reintroduction.

Table 2. Release protocols employed by the Colorado Parks and Wildlife Service to reintroduce Lynx during 1999 and 2000 (Shenk, 2001).

Table 3. Hazard Types and Definitions According to Sainsbury and Vaughan-Higgins (2012) and Masters and Sainsbury (2011).

Table 4. Definitions of traffic light colour categories in terms of the impact level, and requirement for risk assessment and disease mitigation measures.

Table 5. Risk Estimation Matrix

Table 6. Definitions of qualitative likelihoods.

List of Defined Terms

"Advisers" means Clifford Chance LLP and AECOM.

"Annex" means any annex to the Application.

"APHA" means Animal and Plant Health Agency

"Application" means the application by the Lynx Trust [to SNH] for a licence pursuant to section 16 of the WCA to release Lynx into the UK on a trial basis.

"Application Document" means the text of the application only and does not include the annexes to the application.

"DRA" means disease risk assessment

"Infections" means disease causing infectious agents

"IUCN Guidelines" means the Guidelines for reintroductions and Other Conservation Translocations (2013), available online at www.iuchsscrsg.org.

"IUCN Guidelines for DRA" means The IUCN/OIE Guidelines for Wildlife Disease Risk Assessment (2014) available online at

"Key Themes" means the key themes which arise as a result of the Consultations cumulatively as set out in section [] of the National Public Survey, section [] of the National Consultation Report and section [] of the Local Consultation Report.

"Licence" means any licence granted as a result of the Application.

"Lynx" means Eurasian lynx.

"Lynx Trust" means the Lynx UK Trust Community Interest Company with company number 09386570.

"Mitigation" means the measures taken to minimize the risk of Infections and Welfare Hazards in the Lynx and the resident wildlife, pets, livestock and human populations in the release environment.

"NE" means Natural England.

"PALC" means the pre-application local stakeholder and community consultation process as set out in further detail at section [] of the Local Consultation Report.

"Partners" means [the University of Cumbria, the Rewilding Foundation, the White Dog Fund, [insert as we progress]].

"Project" means [•]⁵.

"Project Plan" means [●].

"Release Pen" means [].

"SNH" means Scottish Natural Heritage.

"SNH Code" Translocations & Best Practice Guidelines for Conservation Translocations in Scotland means the Scotlish Code for Conservation.

"Study Area" means the Kielder Study Area and the Eskdalemuir Study Area.

"Swedish Partners" means [●].

"Team" means the Lynx Trust, the Advisers and the Partners.

"Trial" means the proposed trial release of Lynx for a period of [five] years permitted if under a Licence granted pursuant to the Application.

"Trial Animal" means each of the [six to 10] individual Eurasian lynx proposed to be released during the Trial and "Trial Animals" shall be the collective term for them.

"UK" means the United Kingdom.

"WCA" means the Wildlife and Countryside Act 1981.

"Welfare Hazards" means non-infectious agents that can cause disease with negative impacts on the welfare of the Trial Animals

Note to AE: The Common Text uses "Project Plan" and "Project". Do we need to define "Project" or can we just use "Trial"?

1. Introduction⁶

1.1 The Lynx UK Trust CIC

The Lynx Trust is a Community Interest Company (CIC) currently aiming to secure a Licence ("Licence) to conduct a scientific trial reintroduction (population restoration) of Lynx ("Lynx") to the Study Area ("Study Area"), which covers land in Northumberland, the Scottish Borders, Cumbria and Dumfries and Galloway on a time limited basis.

The Lynx Trust ("Lynx Trust") has put together a multidisciplinary team (consisting of its Directors, volunteers, Advisers ("Advisers") and Partners ("Partners")) which comprises academic ecologists, ecological consultants, veterinarians, public consultation experts, economic consultants, socio-economic consultants, forestry specialists, public policy experts, PR experts, media advisers and lawyers. All team members are highly qualified experts in their fields and, as such, the Trust believes that it is perfectly placed to make an Application ("Application") for a Licence to trial the reintroduction of Lynx to England under Section 16 of the Wildlife and Countryside Act 1981 ("WCA").

1.2 The Proposed Project

During the Trial the Team ("Team") would observe, measure and analyse the effects of Lynx on various aspects of the UK's natural, social and economic environments. The ultimate goal of the Trial ("Trial") is to enable [NE/UK Gov/SNH/Scot Gov] to make a final decision as to the desirability of reintroducing Lynx to the wild in the UK on a permanent basis. If such a decision is taken, the Trial Animals ("Trial Animals) and any of their offspring would remain in the wild, acting as the founding population for later generations of British Lynx.

The Project ("Project") includes all preparation required for the Trial and any period after the Trial during which [NE/UK Gov/SNH/Scot Gov] has not made a final decision on Lynx reintroduction. The Project proposal and Project Plan ("Project Plan") is set out in further detail at section [•] [of the Application Document].

This is Annex 8.9 of the Application. The Application is comprised of the Application Document ("Application Document") and [fourteen] supplementary Annexes ("Annexes"), which have been drafted in accordance with the IUCN Guidelines and the Scottish Code, for submission to [NE / SNH]. Where relevant and appropriate, each Annex takes a uniform approach to addressing legislation, policy and guidelines, to presenting and assessing risks and to presenting and addressing the Key Themes of the Consultations.

1.3 Disease Risk Assessment

An important element of the wider Application is a Diseaese Risk Assessment ("DRA") 9 to inform the licensing authorities and the Team of the likelihood of disease associated with the

⁶ AE Comment – Consider splitting below into more introductory sub-headings as in common text.

AE Comment – Insert first para of "The Proposed Project" common text.

⁸ AE Comment – Insert "The Application" common text.

⁹ AE Comment – Definitions are to be included in definitions section. Do not define terms in the body of text.

Trial. Translocation affects host-pathogen communities in the donor and release environments. The primary aim of the DRA is to proactively minimize the likelihood of disease in the Trial Animals, other wildlife, domesticated species and humans, by identifying and assessing the likelihood of disease as a consequence of the Trial and recommending cost-effective disease Mitigation ("Mitigation").

This assessment was completed by a team of veterinary surgeons with extensive experience of the diagnosis, treatment and monitoring of infectious and non-infectious wildlife diseases. Additional information was sourced from the scientific literature using the academic search engines Web of Science and Google Scholar. Expert opinion was sought from an international network of veterinary pathologists, zoo veterinarians, ecologists and epidemiologists with experience of disease monitoring and management in Lynx [or related species such as Canada lynx [Lynx Canadensis]] and their prey species.

[Risk Assessment Common Text Placeholder]¹⁰

1.4 IUCN Guidelines for Wildlife Disease Risk Analysis¹¹

[The IUCN/OIE Guidelines for Wildlife Disease Risk Assessment ["IUCN Guidelines for DRA"] was compiled by the IUCN Species Survival Commission's Wildlife Health Specialist Group, working in concert with the Conservation Breeding Specialist Group, Reintroduction Specialist Group and Invasive Species Specialist Group (4).

The IUCN (2014) recommend that disease risk assessments should be conducted prior to the translocation of species from a donor to a destination environment(4). Historically, DRA frameworks were applied *ad hoc* to situations involving wildlife, often without a consistent approach. Therefore, the IUCN Guidelines for DRA¹² were developed to provide a standardized, evidence based framework to assess the disease risks associated with wildlife management and interventions(4).

The main components of the assessment include the identification of hazards, the assessment of risks associated with the hazards, the management of risks and the communication of outcomes(4). The communication of risk to a wide range of stakeholders (e.g. statutory authorities, scientists, livestock farmers, conservation NGOs) will enable them to make informed decisions regarding the proposed reintroduction and implementation of disease monitoring and Mitigation strategies.

This DRA regarding the Trial is based on the standardized framework described in the IUCN Guidelines for DRA (4)]].¹³

¹⁰ AE Comment – May be required.

AE Comment – Please discuss with GM who is harmonising all legislation/policy discussions across the Application.

AE Comment – Use defined terms throughout.

AE Comment – Consider defining as smaller term?

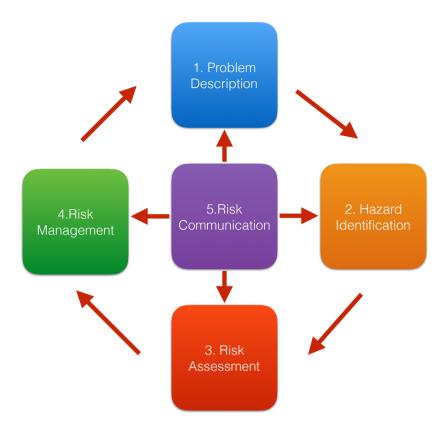


Figure 1. Steps in the DRA (4).

2. Methodology¹⁴

[Quantitative disease risk assessments for the translocation of wild animals are often constrained by insufficient data relating to the prevalence, distribution, transmission and pathogenicity of aetiological agents within the population. This DRA uses a structured qualitative approach to account for the lack of precise quantitative data and to incorporate expert scientific opinion as well as information from the published literature. In qualitative risk assessments the likelihood of the outcome, or the magnitude of the consequences, is expressed in pre-defined terms such as 'high', 'medium' or 'low'.

The authors used the method described by Sainsbury and Vaughan-Higgins (5) which is aligned with the IUCN Guidelines for DRA. Infectious and non-infectious causes of disease were considered to be a hazard if they had crossed an ecological or geographic barrier and were novel to the host. We defined a geographic barrier as a natural or man-made formation or body of water that constrains the physical movement of pathogens or their host. An ecological barrier may not be separated geographically in space but constitutes an unfavorable habitat for a pathogen or its host as a result of competition or the lack of essential resources.

The distinction between a translocation pathway with or without ecological and geographic barriers is paramount, because barriers increase the likelihood of exposure to novel pathogens at the release site. By contrast, translocations between contiguous habitats in the absence of barriers reduces the likelihood of exposure to novel pathogens and the overall risk of disease.

With regard to the Trial, substantial geographic barriers exist between the donor and destination sites. Great Britain has been isolated by sea from Sweden since the land bridge that connected the UK to continental Europe was inundated around 6500BC (6). The North Sea constitutes a geographic barrier of 850kms between the east coast of Great Britain and the West coast of Sweden. Given the extent to which the donor and release countries have been isolated in time and space, it is possible that pathogens carried by the donor Lynx population could constitute a novel disease risk to native wildlife, livestock, pets and humans at the release site. Furthermore, pathogens at the release site could also pose a threat to immunologically naïve donor Lynx.

Diseases that occurred at both donor and release environments were excluded from the DRA unless they occurred as novel subtypes, were notifiable to the local authority in the destination environment or constituted a significant population and/or zoonotic risk. Risk was defined as the probability of a hazardous occurrence and the likely magnitude of the consequences of a deleterious event to animal or human health.

The risk assessment process considered potential disease risk in translocated Lynx and their progeny, and disease risk in other species of wildlife, domesticated livestock and humans at the release site. We compiled an exhaustive list of infectious hazards ["Infections'] and non infectious hazards ["Welfare Hazards'] and considered disease occurrence at every part of the

-

AE Comment – To be harmonised with other risk assessments.

translocation pathway including the donor and release environments and the route of travel. The risk associated with commensal organisms was also evaluated given that translocation related stress could result in host-immunodeficiency and disease. Infectious and non-infectious disease agents were described according to six hazard categories; source, destination, carrier, transport, zoonotic, population (Table 3.).

The assessment of disease risk incorporates the likelihood of disease occurring in the species of concern (Lynx, domesticated livestock, pets, wildlife and humans) and the likelihood and severity of consequences of disease. The overall risk estimation relating to Infections and Welfare Hazards combines the likelihood of disease spread and establishment at the release site with the probable biological, environmental and economic consequences of the disease. The level of risk is categorized as negligible, very low, low, medium or high on the basis of subjective, logical, referenced discussion.

Mitigation recommendations will be described for each hazard unless the overall risk estimation is evaluated as negligible. Mitigation measures against the release and spread of disease might include therapeutic treatments, vaccinations and routine monitoring of Trial Animals.]

3. Demographic Parameters of Founder Population

[The Trial Animals will consist of approximately [6] mature, adult animals sourced from robust populations in Sweden. The translocation of mature adults rather than juvenile or yearling Lynx, ensures the selection of proficient hunters that can exploit the available prey base in the Study Area with an enhanced chance of survival.

Lynx densities are typically 1-3 adults per 100 km2 across Europe, although higher densities of up to 5/100 km2 have been reported from Eastern Europe and parts of Russia (7). However, the natural population density of Lynx varies according to prey abundance and is also limited by the territoriality of individuals.

A typical male home range will overlap with that of two or three females. Therefore, the ratio of females to males in the founder population will be 2:1 to reflect the sex ratio in a typical home range. The release of single sex individuals would result in an increased rate and distance of dispersal as animals search for breeding opportunities. Increased levels of dispersal could compromise the ability to monitor the Trial Animals and increase the anthropogenic risks.

All Trial Animals will receive comprehensive health checks to determine their weight and rule out symptoms of Infections and Welfare Hazards.]¹⁵

-

¹⁵ AE Comment – To be harmonised with Application Document and Project Plan once finalised.

4. Translocation Pathway

4.1 Donor and release sites¹⁶

Discussions with European advisers and senior members of the IUCN Cat Specialist Group have identified Lynx populations in the Baltics, Romania, Slovakia, and Russia. However, The Team have decided to source founder Lynx from Sweden on the basis of reduced disease risks, robust populations and the availability of a surplus hunting quota (8). If the founder population was sourced from a country such as Romania where rabies is endemic, the Lynx would be held in a licensed quarantine facility for a statutory minimum period of four months as stipulated by the Animal and Plant Heath Agency ("APHA"). As Sweden is designated free of rabies, the health of the Trial Animals will not be compromised by the stress associated with a prolonged period of captivity. The Swedish part of the Nordic Lynx population is large and increasing, as Lynx are expanding southwards and have colonized the southern third of the country (8). The most recent census between 2009 and 2011 estimated the national population to be between 1400-1900 individuals (8).

The Team is currently in discussions with a number of potential partner organizations including the Swedish National Veterinary Institute (SVA) and the Swedish Environmental Protection Agency (EPA). These conversations will build reliable partnerships, ensuring proper procedures and safeguards are observed in the identification of potential source populations. The veterinary team will work closely with regional scientific experts to ensure that the removal of donor Lynx does not compromise the ecological and genetic integrity of the source population.

The Team has considered several locations as potential release sites in the UK but has selected Kielder Forest in Northumberland (Figure 2.) for the five-year reintroduction trial (9).

Table 1. Relative benefits of Kielder Forest as a release site for the Trial (9).

Relative advantages of Kielder Forest as a release area
Greater extent of woodland cover
Lower degree of woodland fragmentation
Less farm land within site boundary
Lower density of roads per km ²
Lower number of UK protected species within site boundary
Greater scope for beneficial impacts to the local economy
Fewer potential barriers (rivers, railways, roads)

AE Comment – To harmonise with common text on Site Selection and Application Document of Project Plan on donor selection.



public awareness needs (Table 1.).]

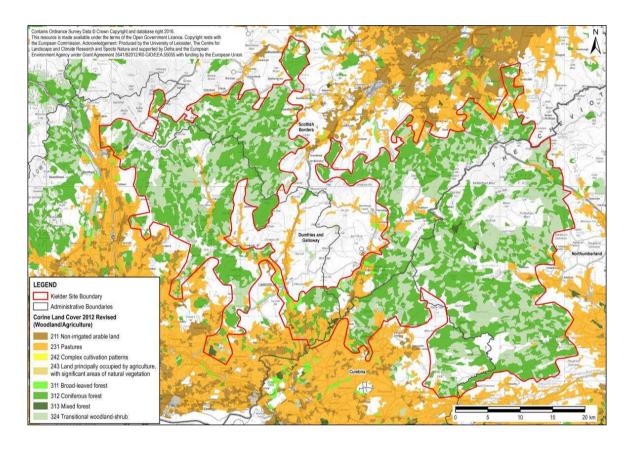


Figure 2. Corine land cover map indicating red boundary of proposed Lynx release area in Kielder Forest (9).

4.2 Capture¹⁷

The Team plans to draw on the expertise of professor Henrik Andren from the Swedish University of Agricultural sciences and others, to adopt best-practice procedures for the capture of Lynx.

Kolbe et al., (10) compared the incidence of capture related injuries in a population (N=63) of Lynx near Seely Lake in Montana using snares, foot hold traps and box traps. The authors observed no injuries in box trapped Lynx but the injury rate for Lynx caught in foot hold traps was 43% and was commonly associated with freezing of the distal extremities as a result of vascular constriction (10). The Norwegian Institute for Nature Research (NINA) published a review of capture related injuries in 140 Lynx trapped between 1995-2007 (11). They observed no injuries in box trapped individuals or in Lynx pursued by dogs and darted in trees. By contrast two out of 48 snare trapped individuals were euthanized as a result of long bone fractures (11).

The Team will prioritize the health and welfare of donor Lynx by employing capture methods that maximize success and minimize stress and trauma.]

4.3 Transport¹⁸

Following capture, donor Lynx will be transferred to IATA approved travel crates and driven to the nearest airport for a direct flight to the UK. The Team will aim to use embarkation and destination airports that are close to the donor and destination sites in Sweden and the UK respectively. The veterinary team will follow expert advice regarding the need for chemical restraint during travel to ensure the health and welfare of animals in transit.

Regulatory compliance regarding the capture and transport of donor Lynx will be achieved through close collaboration between the Team and the Swedish EPA/SVA. Permits will be acquired for the capture, transportation and import of Lynx to the UK, including a CITES permit from the EPA (CITES Appendix II species) and a health certificate issued by Swedish official veterinarians.

4.4 Soft Release¹⁹

There is a substantial body of evidence demonstrating improved survival rates in translocated Lynx following a 'soft release' programme (12). 'Hard release' involves the direct release of animals to the wild from the transit cages. 'Soft release' by contrast restricts the Lynx to an enclosure in which they can shelter and where they receive high quality nutrition. After a predetermined period, the door to the soft release pen is left open to allow individuals to come and go at will, and the provision of food is continued for a number of weeks.

Shenk (12) evaluated release protocols of 96 European Lynx reintroduced to South West Colorado, by the Colorado Parks and Wildlife Service in 1999 and 2000 (Table 2.).

¹⁷ AE Comment – Again for discussion and harmonisation with Application Document and Project Plan.

AE Comment – As above, to harmonise.

AE Comment – As above, to harmonise.

Table 2. Release protocols employed by the Colorado Parks and Wildlife Service to reintroduce Lynx during 1999 and 2000 (12)

Release Protocol	Description
1	Release females as soon as they pass veterinary inspection in Colorado. Release males once females appear to have settled into an area.
2	Release males or females after they have been held in Colorado holding facility for a minimum of 3 weeks and fed a high quality diet.
3	Release males or females after they have been held in Colorado holding facility for a minimum of 3 weeks, fed a high quality diet, and released no earlier than May 1.
3P	Pregnant females released under Protocol 3.
3P?	Possibly pregnant females released under Protocol 3.

The rate of survival during the first 8 months' post-release increased with a minimum holding time of 3 weeks and with a release date no earlier than 01 May (12). An increased mortality rate from starvation was observed post-release in pregnant and juvenile Lynx (12).

Further research relating to the Colorado Lynx reintroduction program (13) revealed that average monthly mortality rates during the first year decreased with time in captivity from 0.205 (95% CI = 0.069, 0.475) for Lynx having spent up to 7 days in captivity to 0.028 (95% CI = 0.012, 0.064) for lynx spending >45 days in captivity before release. No significant additional increases in survival rates were found beyond 5-6 weeks post-release (13).

Despite the additional costs of a soft release program, the Team will follow the available evidence base and release adult, donor Lynx during the spring, after a minimum of 45 days spent in captive release pens. This approach should ensure that the Lynx are in good body condition at the time of release and that they benefit from the seasonal abundance of prey and benign environmental conditions. During their time in captivity, all contact with humans will be kept to a minimum to avoid stress and habituation.

Prior to release, all animals will be fitted with a number of identifying and tracking features. Each Lynx will have a GPS collar fitted and a subcutaneous microchip responder will be implanted.

20

AE Comment – It appears that this is the beginning of methodology of the DRA and the things before it (4 and 5) are summary of Project Plan or intro? Consider separating to 2 sections. 1. Summary of Relevant Project Plans. 2. Methodology.

5. Hazard Categories²¹²²

Table 3. Hazard Types and Definitions According to Sainsbury and Vaughan-Higgins (5) and Masters and Sainsbury (14).

Hazard Type	Definition
Source	Infections carried by translocated individuals from the source environment, which are novel (alien) to the release environment.
Destination	Infections and Welfare Hazards found at the release environment to which the translocated animals are naïve.
Carrier	Those commensal organisms that cause Infections when stressors reduce host immunocompetence and alter the host–parasite relationship.
Transport	Infections that may be encountered during the transport (between the source and destination) which are novel to the translocated animals and/or the release environment.
Zoonotic	Infections carried by the translocated species which can be transmitted to humans and potentially harm the latter.
Population	Infections and Welfare Hazards present at the source and release sites that can potentially have population level effects at the release site.

AE Comment – Possibly to be harmonised with future common text on risk assessment.

AE Comment – Please provide brief text explaining your approach to hazard identification.

6. Hazard Identification²³

We conducted a literature review and sought expert opinion to compile a list of Infections and Welfare Hazards that were carried by Lynx as the definitive or accidental host. We refined the initial list to identify hazards, by filtering those agents that were novel to the host and had crossed an ecological or geographical barrier. Diseases that occurred at both donor and release environments were excluded as hazards unless they occurred as novel subtypes, were notifiable to the local authority in the destination environment or constituted a significant population and/or zoonotic risk.

We considered the potential impacts of each hazard on the Trial Animals, sympatric species of wildlife, livestock, pets and humans at all stages of the translocation pathway, in the absence of mitigation. We employed a traffic light system to display the potential level of severity of each hazard, determine the extent of risk assessment required and ensure that mitigation was proportionate (Table 4).

From an initial list comprising 59 Infections and 4 Welfare Hazards (Appendix 4.) (15 viruses, 13 bacteria, 5 ectoparasites, 19 endoparasites, 6 protozoa, 1 fungi and 4 Welfare Hazards) we assigned 5 Infections (rabies virus, *Fransicella tularensis*, *Sarcoptes scabei*, tick-borne encephalitis virus and *Echinococcus multilocularis*) and 2 Welfare Hazards²⁴ (illegal persecution, generic stress) to the red colour code category.

Table 4. Definitions of traffic light colour categories in terms of the impact level, and requirement for risk assessment and disease mitigation measures.

Colour Categories	Definition of Traffic Light Categories
Red	Severe impacts at an individual or population level Full risk assessment recommended Mitigation measures essential
Amber	Moderate impacts at an individual or population level Partial risk assessment recommended Mitigation measures advisable
Green	Negligible impacts at an individual or population level Risk assessment not required Mitigation measures not required

-

AE Comment – Possibly to be harmonised with future risk assessment common text.

AE Comment – Consider using as a defined term.

7. Risk Assessment²⁵

The risk assessment follows the identification and categorization of individual Infections and Welfare Hazards and describes the biological pathways for each hazard from the acquisition of disease at the donor site to the release and spread at the release site.

The assessment of disease risk incorporates the likelihood of Lynx carrying and releasing Infections at the recipient site (release assessment), the probability of disease transmission to Lynx and sympatric, resident species at the release site (wildlife, domestic livestock, pets and humans) (exposure assessment) and the likelihood and severity of biological, environmental and economic consequences of the disease (consequence assessment). The overall qualitative risk estimation (Table 5.) for Infections and Welfare Hazards combines the results of the release, exposure and consequence assessments. The definitions of qualitative likelihoods that are used in the Risk Estimation Matrix (Table 5.) (negligible, very low, low, medium or high) were established on the basis of subjective, logical, referenced discussion (Table 6.)

Table 5. Risk Estimation Matrix

Likelihood of
0
<u> </u>
f release and exposure
=
6
release
е
an
⊇
0
ტ
Ã
×
Š
⊆
ď

High	Negligible risk	Very low risk	Low risk	Moderate risk	High risk
Moderate	Negligible risk	Very low risk	Low risk	Moderate risk	High risk
Low	Negligible risk	Negligible risk	Very low risk	Low risk	Moderate risk
Very Low	Negligible risk	Negligible risk	Negligible risk	Low risk	Moderate risk
Negligible	Negligible risk	Negligible risk	Negligible risk	Negligible risk	Negligible risk
	Negligible	Very low	Low	Moderate	High

Likelihood and severity of consequences

_

AE Comment – As above, possibly to be harmonised with Future Risk Assessment Common Text.

Table 6. Definitions of qualitative likelihoods.

Likelihood	Descriptive definition
High	The event is very likely to occur
Moderate	The event will occur with an even probability
Low	The event is unlikely to occur
Very Low	The event is very unlikely to occur
Negligible	The event will almost certainly not occur

7.1 Release, exposure and consequence assessments

Release Assessment

Release in this context relates to the introduction into the environment or direct transmission to another individual, of an Infection carried by the Trial Animals. Release assessments are not required for destination hazards or for source/transport/carrier/zoonotic hazards where the likelihood of release is negligeable. Release assessments do not apply to Welfare Hazards.

Exposure Assessments

Exposure assessments evaluate the risk of disease transmission from Lynx to sympatric resident species at the release site and vice versa. For Welfare Hazards, exposure assessments will determine the susceptibility of a species acquiring or encountering the hazard.

Consequence Assessment

Consequence assessment determines the severity and likelihood of biological, environmental and economic consequences given the release and exposure of Infections and Welfare Hazards.

8. Mitigation Strategy²⁶

The Mitigation ("Mitigation") strategy is designed to provide an overview of the measures recommended for implementation at each stage of the translocation pathway, to minimize the risk of Infections and Welfare Hazards in the founder Lynx and the resident wildlife, pets, livestock and human populations in the release environment. The strategy will focus on red colour-coded hazards with the potential for severe impacts at an individual or population level. Hazard-specific recommendations will include management measures, diagnostic testing and preventative healthcare treatments, but it is beyond the scope of this document to provide details of pharmacological products and dosages.

8.1 Pre-Export

Following capture in Sweden and prior to export, thorough non-invasive clinical observations of the Lynx will be conducted at regular intervals in the holding facility. These observations should be conducted during the day and at night using a thermal scope to account for their nocturnal activity patterns. The veterinary team will evaluate the physical condition, general demeanor, appetite and thirst, mobility and behaviour of the Lynx. Furthermore, observations will identify hazard specific symptoms such as pruritis (*Sarcoptes scabei, Otodectes cynotis*) and evidence of stress induced trauma to distal extremities. The detection of clinical disease symptoms will, at the discretion of the veterinary team, result in the provision of therapeutic treatments pre-transport, or the removal of the individual from the translocation project.

Prior to travel, a general anaesthetic will be administered to conduct a physical examination, collect biological samples for diagnostic screening, apply prophylactic and therapeutic treatments and take morphometric measurements. To mitigate the risks of morbidity and mortality associated with anaesthesia, the veterinary team will employ best practice protocols devised by Norwegian School of Veterinary Science in Tromsø (15). Routine checks will include cardiac auscultation and otoscopic/opthalmoscopic examination of the ears and eyes respectively. Examination of the skin will identify dermatological lesions and evidence of ectoparasites and palpation/manipulation of the bones and joints will reveal musculoskeletal abnormalities such as traumatic injuries.

Biological samples including blood, urine, tissue, faeces and conjunctival/oropharyngeal swabs will be taken for immediate analysis at the Swedish National Veterinary Institute (SVA) and for storage at -80°C to enable retrospective testing as required. Serum and EDTA samples will be analyzed for routine biochemistry and hematology and specifically for *Sarcoptes scabei, Francisella tularensis* and Tick-borne encephalitis virus (TBEV) antibodies by enzymelinked immuno-sorbent assay (ELISA). Blood smears will be examined for haemoparasites and skin scrapes will be assessed microscopically for evidence of *Sarcoptes scabei*. Faecal analysis will include routine microscopy for endoparasites, routine culture for pathogenic bacteria, as well as hazard specific coproantigen ELISA testing for *Echinococcus multilocularis*.

_

AE Comment – TBD – would it make sense to cover mitigation after risks or perhaps in the same table as risks, including "residual risks"?

Prophylactic interventions administered prior to export comprise vaccinations and treatments against endoparasites and ectoparasites. Only inactivated vaccines should be utilized, to prevent vaccine-induced disease and post-vaccine shedding. Despite the low level of risk associated with feline leukaemia virus (FeLV), feline calicivirus (FCV), feline herpesvirus (FHV), feline parvovirus (FPV) and canine distemper virus (CDV) in Lynx, vaccinations should be considered to reduce transmission from feral cats and foxes in the destination environment. Anthelmintics with proven efficacy against *Echinococcus multilocularis* should be given and ectoparasiticides should be effective against *Sarcoptes scabei* as well as ticks, fleas and lice. Therapeutic treatments will only be administered on a case by case basis as and when required.

8.2 Transport

The management and mitigation of transport related stress is discussed in the 'Generic Stress' risk assessment in Appendix 2. To avoid the transport of fomites and vectors that could carry Infections, biosecurity measures must be maintained to the highest standards in transit from Sweden to the UK. IATA compliant travel crates should be disinfected pre-travel with a non-toxic, viricidal/bacteriocidal disinfectant that meets all regulatory and accreditation standards and is DEFRA approved. Particular attention should be paid to eliminating arthropod vectors pre-travel and the travel crates should not be in the proximity of other animals during transport.

8.3 Release Enclosures

Strict biosecurity standards must be adhered to in the soft-release enclosures. These include the disinfection of all building materials used in the construction of enclosures and the regular disinfection of utensils. Wild prey such as roe deer should be provided to mimic the natural diet to which the Lynx were accustomed in the source environment. Veterinary staff will conduct meat inspections prior to feeding wild prey to identify lesions associated with infectious disease such as *Mycobacterium bovis* and *Sarcoptes scabei*. The provision of fresh or frozen carcasses will help to prevent bacterial contamination with pathogens such as Salmonella and Campylobacter species. If flowing water is not available in the release enclosure, standing water sources should be refilled on a regular basis. Cage design should exclude cats and dogs from the enclosures to avoid exposing the Lynx to common canine and feline diseases such as FeLV, FIV, FCH and FHV. Enclosure management plans should advise against the Lynx Team bringing cats and dogs anywhere near the enclosure compound.

The enclosures should be managed by experienced feline handlers who will adhere to a strict cleaning and maintenance rota. Regular clinical observations to monitor the health and welfare of the Lynx, should be implemented during the day and night as stipulated in the pre-export period. Therapeutic interventions will only be carried out on a case by case basis if symptoms of disease have been identified. Repeated non-invasive faecal testing will help to monitor parasite loads and could be used to evaluate stress through the analysis of faecal glucocorticoid metabolite (FGM) levels.

Prior to release a second anaesthetic will be administered, to enable a comprehensive physical examination and weight check to be carried out and to fit individual Lynx with satellite collars. The detection of clinical disease symptoms will, at the discretion of the veterinary team, result in the provision of therapeutic treatments pre-release or the removal of the individual from the translocation project.

8.4 Post-release

Following release, the Lynx will be satellite tracked in real time to monitor their movements and alert farmers of the proximity of Lynx to livestock, as discussed in the risk assessment for 'Illegal Persecution' in Appendix 2. It is imperative that the community engagement programme implemented prior to the licensing stage is continued in the release zone post-licensing. This will help to inform the community of the mitigation measures that have been applied to protect local wildlife, domestic pets and livestock and promote the peaceful coexistence of humans and Lynx in the Kielder Forest.

Despite the challenges of monitoring the health of Lynx in the post-release period, camera trap images and non-invasive scat analysis will provide some information regarding the health status and dietary preferences of the founder Lynx population.

8.5 Post-mortem examination

Lynx mortality events will be investigated at all stages of the translocation pathway by Dr Julian Chantrey from the University of Liverpool Veterinary Institute according to the necropsy protocol outlined in Appendix 3. Diagnostic tests will include gross necropsy, histopathology and toxicology screens and the cause of death will inform the mitigation strategy and assist in protecting the remaining founder Lynx population. Necropsy examinations will also extend to any large prey species (roe deer, fox) that are found dead in the proximity of the soft release enclosures.

9 Consultations²⁷

-

AE Comment – Insert section from Common Text as to how Key Themes have been addressed.

10 Discussion

This DRA is intended for use by all parties involved in the proposed Lynx reintroduction Project including the Lynx Trust Team as well as the statutory authorities (NE, SNH,APHA) and Swedish Partners ("Swedish Partners"). The most labour-intensive and financially costly interventions are largely concerned with the Swedish stages of the programme. Interventions at UK release pens are less onerous and rely partially on clinical observation and animal and enclosure management skills of the staff involved.

Our mitigation strategy was formulated in the light of the risk assessment process which has drawn on a reference library of published articles relating in some part to feline diseases. There were many instances where the certainty level of our risk assessments was compromised by lack of disease surveillance data both in Sweden and UK, and lack of data on disease in wild felids, leading to assumptions relating to predicted prevalence and/or Lynx disease susceptibility.

The UK government's regulatory APHA is mainly concerned with the introduction of exotic Swedish Infections resulting from the Trial, however the threat of these Infections to the UK is greatly diminished by the infrequent and sporadic nature of all Infections for which the Lynx is an accidental host. The largely solitary nature of the lynx and overall low population densities at which it lives, even in optimal habitat, mean Infections are relatively rare in this species.

The threat of endemic UK disease to the newly introduced Trial Animals is of lesser concern for the UK animal health authorities but is difficult to accurately assess due to many unknowns. As an example, sarcoptic mange is endemic at low levels in the fox population in Kielder. Lynx can contract this parasitic disease from fox prey and may be clinically affected, however British mange strains may differ in virulence from mainland European types. This could hold true for a range of endemic UK infections reservoired in wildlife species eg. hantavirus, louping ill virus, Anaplasma spp, cowpox virus, *Borrelia burgdorferi* etc however, the likelihood is their virulence is not significantly different between the two countries.

Without disease specific Mitigation measures, Infections could reduce the survival rates and reproductive potential of the Trial Animals. However The DRA findings indicate that the five red coded pathogens, can all be mitigated by simple procedures such as selecting the lynx from disease free areas (Sweden for rabies); treating all imported lynx with anthelmintics (Echinococcus) or acaricide drugs (TBEV, Tularaemia, mange) or a combination of these factors (Echinococcus/ Tularaemia). Therefore it is likely that anthropogenic Welfare Hazards such as illegal persecution will pose a greater threat to the Lynx than Infections acquired along the translocation pathway¹⁶.

11 Acknowledgements

The veterinary team acknowledge with thanks the assistance and expertise provided by the following organisations and individuals in compiling this DRA report:

- Animal and Plant Health Agency (APHA)
- Dr Daniel R. Stahler, Yellowstone National Park
- Dr John Lewis MA, VetMB, PhD, MRCVS, International Zoo Veterinary Group (IZVG)
- Dr Tony Sainsbury Institute of Zoology, Zoological Society of London
- Hanna Dittrich Soderman and Klas Allander, Swedish Environmental Protection Agency (EPA).
- Institute of Veterinary Science, University of Liverpool
- Professor Henrik Andren, Swedish University of Agricultural sciences
- Swedish National Veterinary Institute (SVA)
- Tim Jagger BVM&S, MSc, FRCPath, MRCVS, Idexx Laboratories Ltd.
- University of Cumbria

12 References

- 1. CD (Council Directive) 92/43/EEC. (1992) The conservation of natural habitats and of wild fauna and flora.
- 2. IUCN/SSC (2013). *Guidelines for Reintroductions and Other Conservation Translocations. Version 1.0.* Gland, Switzerland: IUCN Species Survival Commission, viiii + 57 pp.
- 3. National Species Reintroduction Forum (2014) *Best Practice Guidelines for Conservation Translocations in Scotland Version 1.1.* Scottish Natural Heritage.
- 4. World Organisation for Animal Health (OIE) & International Union for Conservation of Nature (IUCN) (2014) Guidelines for Wildlife Disease Risk Analysis. OIE, Paris, 24 pp. Published in association with the IUCN and the Species Survival Commission.
- 5. Sainsbury, A.W. and VAUGHAN-HIGGINS, R.J. (2012) Analyzing disease risks associated with translocations. *Conservation Biology*, *26*(3), pp.442-452.
- 6. Coles, B.J. (1998) Doggerland: a speculative survey. Proceedings of the Prehistoric Society 64, 45-81.
- 7. Breitenmoser-Würsten, C., Zimmermann, F., Ryser, A., Capt, S., Laass, J., Siegenthaler, A. & Breitenmoser, U. (2001) *Untersuchungen Zur Luchspopulation in Den Nordwestalpen der Schweiz 1997–2000*. KORA Bericht 9d, Switzerland.
- 8. Kaczensky, P., Chapron, G., von Arx, M., Huber, D., Andrén, H., Linnell, J.D.C. (2012) Status, Management and Distribution of Large Carnivores Bear, Lynx, Wolf & Wolverine in Europe. European Commission, Brussels.
- 9. White, C., Eagle, A., O'Donoghue, P., Rowcroft, P. & Wade, M. (2016), 'Reintroduction of the Eurasian Lynx to the United Kingdom: Trial site selection', Prepared for the Lynx UK Trust by AECOM.
- 10. Kolbe, J.A., Squires, J.R. and Parker, T.W. (2003) An effective box trap for capturing lynx. *Wildlife Society Bulletin*, pp.980-985.
- 11. Odden, J., Linnell, J.D.C., Arnemo, J. M. & Berntsen, F. (2007) Refinement of research capture techniques capture of Eurasian lynx in Norway 1995–2007. NINA Minirapport 203.
- 12. Shenk, T. M. (2001) Post-release monitoring of lynx reintroduced to Colorado. Annual progress report for the USA Fish and Wildlife Service. Colorado Division of Wildlife, Denver, USA.
- 13. Devineau, O., Shenk, T.M., Doherty, P.F., White, G.C. and Kahn, R.H. (2011) Assessing release protocols for Canada lynx reintroduction in Colorado. *The Journal of Wildlife Management*, 75(3), pp.623-630.
- 14. Masters, N., Sainsbury A.W. (2011) Disease risk analysis for the wild to wild translocation of the smooth snake within the UK. Zoological Society of London and Natural England. pp 62
- 15. Arnemo, J., Evans, A. and Fahlman, A. (2007) *Biomedical protocols for free-ranging brown bears, gray wolves, wolverines and lynx. Norwegian School of Veterinary Science*. General Technical Report, Tromsø, Norway.
- 16. Schmidt-Posthaus, H., Breitenmoser-Wörsten, C., Posthaus, H., Bacciarini, L. and Breitenmoser, U., (2002) Causes of mortality in reintroduced Eurasian lynx in Switzerland. *Journal of Wildlife Diseases*, *38*(1), pp.84-92.

13. Appendices²⁸

Appendix 1. **Individual Disease Risk Assessments: Infections**

A) Rabies Virus	Hazard Categories (Zoonotic/Source/Population)
Justification for Hazard Status	Rabies is an encephalitis caused by rabies virus, a member of the Rhabdoviridae family. It is an acute zoonotic viral infection that is almost invariably fatal in humans once symptoms develop. Transmission is generally through the bite of an infected animal, usually dogs but also cats, bats and other wildlife (1). In humans, a course of rabies vaccination can prevent infection and death. The European distribution of rabies is in the far east with endemic foci being present in Turkey and former Eastern bloc countries (2). Two epidemiological life cycles exist, sylvatic rabies for which the red fox (<i>Vulpes vulpes</i>) is the wildlife reservoir and urban rabies which is present in dogs (3). Both types may occur in an outbreak but urban canine rabies is by far more important for public health and contributes to 99% to the human death toll (2). Transmission of the virus can also occur to a wide variety of other mammalian species, including cats.
Risk Assessment	
Release Assessment	The UK (since 1922) and Sweden (since 1886) are categorised as being free of rabies (4), as is Finland, so the nearest overland sites of endemic rabies infection from NE Sweden are in north western Russia (over 250km distant) (2).
	In Europe, the main reservoir host species for rabies is the red fox which acquires infection by fighting or saliva contact with infected animals. The susceptibility of animals to rabies varies depending on the species but felids are only moderately susceptible (5). Furthermore, cats usually get infected through perforating bite wounds rather than through ingestion of an infected prey item (6). Although there is no documented evidence of rabies infection in Swedish lynx (7), sporadic cases of rabies have been diagnosed in lynx elsewhere. In Slovakia, Fernex (8) identified six of a thousand (0.6%) lynx caught or killed in a ten-year

²⁸ AE Comment – To be harmonised with other risk assessment approaches.

	period had rabies. All six showed the paralytic signs of the disease (dumb rabies), with absence of aggression, however, a case of rabid lynx showing aggression (furious rabies) has also been described once (9).
	Thus with very low rates of infection, a low density population made up of solitary individuals and less susceptibility to infection compared to foxes, the Lynx is not thought to play a significant role in the epidemiology of rabies in Europe.
Exposure Assessment	Rabies virus is only present in the saliva or CNS tissue of the infected animal and once outside the host the virus is inactivated by high temperatures (>21oC), desiccation and exposure to sunlight (1) but it can remain infectious for extended periods at low or freezing temperatures (3). In carnivores, wild canids are considered to be easily infected, whereas cats are much less susceptible (10). Rabies incubation times depend on viral quantity, virulence and location of the inoculation site but the average is 2 to 8 weeks. The virus may be present in saliva from an infected animal up to five days before the outbreak of clinical symptoms (9).
Consequence Assessment	The release and exposure of rabies virus in the UK could present a risk to public health and would have environmental and economic consequences as a result of the necessity for dogs to be vaccinated against rabid foxes. The increased risk of humans contracting rabies through handling of dogs (or foxes) may occasionally lead to clinical cases and necessitate human vaccination, treatment and hospitalization (11). The mitigation of rabies in wildlife populations would require perimeter vaccination of foxes around the release site by the Department of Agriculture, Environment and Rural Affairs (DEFRA) within and beyond the Kielder area, however, vaccination has proven highly effective in eliminating rabies from fox populations in western Europe (12). Despite the significant nature of the consequences, the low probability of occurrence, release and exposure
Overall Risk Estimation	all reduce the likelihood of serious consequences occurring. Very low.
Risk Management	The statutory UK 4 month quarantine period either at the release site or in Sweden will outlast the incubation period so disease would occur in and be confined to quarantine facilities. In addition, DEFRA may grant quarantine exemption given the source country is rabies free.

- 1. CDC https://www.cdc.gov/rabies/exposure/materials.html
- 2. WHO https://www.who-rabies-bulletin.org/Journal/Article_Search.htm
- 3. OIE http://www.oie.int/fileadmin/Home/eng/Animal Health in the World/docs/pdf/Disease cards/RABIES FINAL.pdf
- 4. UK Government PHE https://www.gov.uk/government/publications/rabies-risks-by-country/rabies-risks-in-terrestrial-animals-by-country/s
- 5. Artois, M., Bourhy, H., Mueller, T. F., Selhorst, T., & Smith, G. (2012). Lyssavirus infections. In D. Gavier-Widen, P. Duff, & A. Meredith (Eds.), *Infectious Diseases of Wild Mammals and Birds in Europe* (First., pp. 86–97). Blackwell Publishing Ltd., UK.
- 6. Bell J and Moore G 1971. Susceptibility of carnivores to rabies virus administered orally. American Journal of Epidemiology 93, 176-182
- 7. Malmsten J and E Agren 2014. Wildlife disease monitoring in Sweden. National Veterinary Institute, SVA, Uppsala ISSN 1654-7098
- 8. Fernex M 1976 Impact du lynx sur l'epizootie de rage vulpine. Bulletin de la Societe Industrielle de Mulhouse 770, 97-98
- 9. Kolar 1976 Tollwut bei einem Karpathenluchs (*Lynx lynx*) Verhandlungsbericht des XVIIII. Internationalen Symposiums uber die Erkrankungen der Zootierer, pp 103-104
- 10. Brömel, J 1994. Tollwut. In: Wildhygiene, J. Dedeck and T. Steineck (eds). Gustav Fischer Verlag, Jena, Germany. Pp. 107–108.
- 11. UK Government PHE https://www.gov.uk/government/collections/rabies-risk-assessment-post-exposure-treatment-management
- 12. Aubert M et al. 1995 Bull Acad Natl Med. May;179(5):1033-54. Epidemiology and campaign against rabies in France and in Europe.

B) Tick borne encephalitis virus	Hazard Categories (Zoonotic/Source/Population)
Justification for Hazard Status	Tick-Borne Encephalitis (TBE) is a viral disease caused by a member of the Flavivirus family. The European distribution of TBE is in central and northern Europe, and in Sweden, the risk is present throughout the country but particularly in the south east (3). It is an acute zoonotic viral infection that occasionally causes serious neurological disease in humans, however, the vast majority of those who are infected will have mild or no symptoms. Ticks act as both the vector and reservoir for TBE and transmission is generally through the bite of an infected <i>Ixodes ricinus or Ixodes persulcatus</i> (1). The main hosts are small rodents but larger animals (eg. deer) although serving as feeding hosts for the ticks, do not play a role in maintenance of the virus. Humans are accidental hosts (4) and the European human case fatality rate is less than 2% but about 46% of diagnosed patients suffer permanent neurological sequelae [2]. Person-to-person transmission has not been reported with the exception of vertical transmission, from an infected mother to foetus (4). Louping ill virus (LIV) is a related member of this Flavivirus family and is endemic in sheep and hares in the UK, and other parts of Europe (7). It causes disease primarily in sheep and has been reported as the cause of a TBE-like illness in people (4). Unlike TBE, LIV causes viraemia and fatal encephalomyelitis in domesticated animals when infected ticks feed on them (5).
Risk Assessment	T
Release Assessment	There are no scientific records of Lynx being infected with TBE, nor being exposed from positive serology results. Larger species, like deer, are dead-end hosts for the virus (Carpi 2008) so it is probable that Lynx also act similarly, acting as accidental hosts. The only documented native UK species rodent species in Kielder forest capable of allowing TBE to replicate is the bank vole (<i>Myodes glareolus</i>)(10). As the TBE virus is transovarially transmitted (9), the risk is importing Lynx which have feeding adult ticks which can then reproduce, fall off and these surviving larvae parasitise UK bank voles, so re-establishing the TBE infection cycle. The Lynx is not thought to play a significant role in the epidemiology of TBE in Europe.
Exposure Assessment	The main vector for TBE virus (TBE) transmission to humans in Sweden is the nymph stage of the common tick, <i>Ixodes ricinus</i> . The main mode of transmission and maintenance of TBE in the tick population is considered to be when infective nymphs co-feed with uninfected but susceptible larvae on rodents (within the UK this would be the bank vole). In wildlife, the roe deer, <i>Capreolus capreolus</i> is the main host for the reproducing adult <i>I. ricinus</i> ticks (11). Ixodes ricinus ticks can feed on Lynx but this is less common than deer (12).

Consequence Assessment	The release and exposure of TBE virus in the UK could present a risk to public health and would have economic and environmental consequences as a result of the infection becoming established within the UK bank vole population. The increased risk of humans contracting TBE through being fed on by infected ticks could lead to clinical cases and necessitate vaccination and treatment. Despite the serious and intractable nature of TBE infection, the low probability of occurrence, release and exposure all reduce the likelihood of serious consequences occurring.
Overall Risk Estimation	Moderate.
Risk Managment	The mitigation of TBE virus in wildlife populations is difficult. Preventative measures such as vaccination of forestry workers and farmers in the local Kielder area have proven efficacy in protecting the target human susceptible population in Sweden (11). Acaricide treatment such as ivermectin and screening of the Lynx post-capture and pre-travel to the UK should prevent any TBE infected ticks being imported into the UK.

- 1. Å Lundkvist, A Wallensten, S Vene, M Hjertqvist Tick-borne encephalitis increasing in Sweden, 2011 Eurosurveillance, Volume 16, Issue 39, 29 September 2011
- 2. Haglund M, Gunther G. Tick-borne encephalitis--pathogenesis, clinical course and long-term follow-up. Vaccine. 2003;21 Suppl 1:S11-8.
- 3. WHO http://www.who.int/immunization/topics/tick_encephalitis/en/
- 4. CDC https://www.cdc.gov/vhf/tbe/
- 5. K. L. Mansfield, N. Johnson, L. P. Phipps, J. R. Stephenson, A. R. Fooks and T. Solomon Tick-borne encephalitis virus a review of an emerging zoonosis Journal of General Virology (2009), 90, 1781–1794
- 6. Dobler G. Zoonotic tick-borne flaviviruses. Veterinary Microbiology 140 (2010) 221–228
- 7. M.Laurenson, R Norman, L. Gilbert, H Reid and P Hudson. Journal of Animal Ecology 2003 Vol 72, Issue 1, pages 177–185. Identifying disease reservoirs in complex systems: mountain hares as reservoirs of ticks and louping-ill virus, pathogens of red grouse
- 8. Carpi, G., Cagnacci, F., Neteler, M., & Rizzoli, A. (2008). Tick infestation on roe deer in relation to geographic and remotely sensed climatic variables in a tick-borne encephalitis endemic area. *Epidemiology and infection*, 136(10), 1416-1424.
- 9. B Danielová, V., Holubová, J., Pejcoch, M., & Daniel, M. (2002). Potential significance of transovarial transmission in the circulation of tick-borne encephalitis virus. *Folia Parasitologica*, *49*(4), 323-325.
- 10. Labuda, M., Kozuch, O., Zuffová, E., Elecková, E., Hails, R. S., & Nuttall, P. A. (1997). Tick-borne encephalitis virus transmission between ticks cofeeding on specific immune natural rodent hosts. *Virology*, *235*(1), 138-143.
- 11. Jaenson, T. G., Hjertqvist, M., Bergström, T., & Lundkvist, Å. (2012). Why is tick-borne encephalitis increasing? A review of the key factors causing the increasing incidence of human TBE in Sweden. *Parasites & vectors*, 5(1), 184.
- 12. M.P. Degiorgis, A.-L. Berg, C. Hård af Segerstad, T. Mörner, M. Johansson and M. Berg. Borna Disease in a Free-Ranging Lynx (Lynx lynx) J. Clin. Microbiol. August 2000 vol. 38 no. 8 3087-3091

C) Echinococcus	Hazard Categories (Zoonotic/Source/Population)
multilocularis	
Justification for Hazard Status	Alveolar echinococcosis caused by <i>Echinococcus multilocularis</i> (EM) constitutes one of the most pathogenic zoonoses in temperate and arctic areas of the Northern hemisphere (1). Human infection is acquired through the ingestion of eggs excreted in the faeces of the definitive host and is characterized by a prolonged incubation period of up to 15 years followed by a potentially fatal infiltrative tumour-like disease in the liver and other associated organs (2). The UK is currently listed as free of EM and DEFRA conducts routine surveillance of fox carcasses in the UK (3). Prior to 2010, EM had never been detected in Sweden, but following the discovery of infected foxes in Denmark, a national monitoring programme was established based on analysis of fox scat to determine the prevalence and distribution of EM in Sweden (4). In 2011, one infected fox was diagnosed in South-West Sweden (Västra Götaland County). During 2012-2014, active monitoring of shot foxes was continued by the board of agriculture and three positive cases were diagnosed from 2779 fox scat samples in the municipalities of Uddevalla, Katrineholm, and Gnesta (4). Furthermore, a project led by the University of Agricultural Sciences (SLU), to determine the prevalence of EM in rodent intermediate hosts confirmed one case of EM in a water vole (Arvicola species) (4). EM is considered to be endemic with a low prevalence in Sweden.
Risk Assessment	
Release Assessment	The most common definitive host for EM is the red fox (<i>Vulpes vulpes</i>), although sexual reproduction of the parasite can also occur in other members of the family canidae (4). Foxes acquire the infection by ingesting rodent intermediate hosts. Although there is no evidence of EM infection in Swedish lynx (2), cats can act as definitive hosts under natural and experimental conditions (5,6,7). In Europe EM has been diagnosed in cats from Baden-Württemburg, Germany (6), and the Saubian Alps, Germany (5). Vogel (1957) succeeded in infecting 5 of 6 cats with EM from southern Germany, but noted that felines were poor hosts on the basis that they produced smaller worms and fewer eggs. The definitive hosts of EM remain asymptomatic so there is a low likelihood that untreated Lynx could contaminate the destination environment by carrying infection and releasing eggs in their stools.
Exposure Assessment	The eggs of EM are rapidly killed by high temperatures and desiccation, but experimental evidence from South West Germany established a maximum survival time of 240 days under benign environmental conditions during the autumn and winter (8). Eggs shed in the environment could infect rodent intermediate hosts and result in the transmission of EM to red foxes. Furthermore, animal handlers including scientists and wildlife veterinarians could contract EM by handling infected Lynx during capture, transport and disease monitoring. However, the

	likelihood of exposure remains low as felines only constitute accidental hosts for EM and the disease has never been diagnosed in Swedish lynx (2).
Consequence Assessment	The release and exposure of EM to the UK could pose a serious risk to public health and would have economic and environmental consequences. The increased risk of humans contracting alveolar echinococcosis could lead to clinical cases and necessitate expensive treatments including surgical resection of hydatid cysts in the liver and long term chemotherapy. The circulation of the parasite in fox and rodent populations would require intense surveillance of fox scat/carcasses by DEFRA within and beyond the Kielder area. Despite the severe nature of the consequences, the low probability of release and exposure also reduces the likelihood of consequences occurring.
Overall Risk Estimation	Low
Risk Management	Praziquantel (5mg/kg) has proven efficacy against EM and will be administered to all Trial Animals prior to treatment faecal testing for coproantigen (CoproAntigen ELISA) will be carried out on all Lynx and ELISA positive cases will be examined using sedimentation and counting techniques.

- 1. Kern, P., Bardonnet, K., Renner, E., Auer, H., Pawlowski, Z., Ammann, R.W., Vuitton, D.A. and Kern, P., 2003. European echinococcosis registry: human alveolar echinococcosis, Europe, 1982-2000. *Emerging infectious diseases*, *9*(3), pp.343-349.
- 2. Surveillance of infectious diseases in animals and humans in Sweden 2014, National Veterinary Institute (SVA), Uppsala, Sweden. SVA:s rapportserie 31 ISSN 1654-7098. Available at: http://www.sva.se/globalassets/redesign2011/pdf/om sva/publikationer/surveillance2014.pdf. (Accessed: 12.02.17).
- 3. DAERA (2016) Echinococus multilocularis. Available at: https://www.daera-ni.gov.uk/articles/echinococcus-multilocularis. (Accessed: 08.01.17).
- 4. Wildlife disease monitoring in Sweden (2014) National Veterinary Institute, SVA, Uppsala, Sweden. SVA's report series 33, ISSN 1654-7098. Available at: http://www.sva.se/globalassets/redesign2011/pdf/om sva/publikationer/vilda-djur/wildlife-disease-monitor-2014.pdf. (Accessed: 18.12.16)
- 5. Zeyhle, E. and Bosch, D., 1982. Comparative experimental infections of cats and foxes with Echinococcus multilocularis. *Zentralbl. Bakteriol. Parasitenkd*, 277, pp.117-118.
- 6. Eckert, J., Müller, B. and Partridge, A.J., 1974. The domestic cat and dog as natural definitive hosts of Echinococcus (Alveococcus) multilocularis in Southern Federal Republic of Germany. *Tropenmedizin und Parasitologie*, *25*(3), pp.334-7.
- 7. *Vogel, H.:* Uber den Echinococcus multilo- cularis Siiddeutschlands. I. Das Bandwurm- stadium von Stammen menschlicher und tierischer Herkunft. Z.Tropenmed.Parasi- tol.8 (1957) 404-454
- 8. Veit, P., Bilger, B., Schad, V., Schäfer, J., Frank, W. and Lucius, R., 1995. Influence of environmental factors on the infectivity of Echinococcus multilocularis eggs. *Parasitology*, *110*(01), pp.79-86.

D) Sarcoptes scabei	Hazard Categories (Zoonotic/Population/Carrier)
Justification for Hazard Status	Sarcoptic mange is a highly contagious epizootic disease caused by the <i>Sarcoptes scabei</i> , a submacroscopic burrowing skin mite (1). Infection has been recorded in 104 wild and domestic species but is most prevalent in red foxes (<i>Vulpes vulpes</i>) and grey wolves (<i>Canis lupus</i>) in North America and red foxes and dingoes (<i>Canis familiaris dingo</i>) in Australia (2).
	Sarcoptic mange was unrecorded in Scandinavia until 1967 when it was diagnosed in a red fox in the South West of Finland and in a Swedish red fox in 1972 (3). The subsequent spread of <i>Sarcoptes scabei</i> in Sweden was determined by the population dynamics of the red fox and resulted in the first positive diagnosis in Lynx in 1980 (3). Sarcoptic mange now constitutes the most common infectious disease of Lynx in Europe, and is characterized by chronic dermatological lesions culminating in emaciation, dehydration, cachexia and a high mortality rate (4,1). In 2014 the SVA diagnosed 10 cases of sarcoptic mange from 77 fallen Lynx, as part of their annual infectious disease monitoring programme (4). Sarcoptic mange is widespread in The UK with the highest estimated prevalence in central and southern England and a reduced incidence in the North of England and Scotland (5). Human scabies does not constitute a major public health concern as zoonotic infections are commonly short term, self limiting and characterized by mild dermatological symptoms (1).
Risk Assessment	
Release Assessment	Sarcoptic mange has a protracted incubation of 10-72 days and a prolonged disease period to death (6). Furthermore, the ectoparasite can survive in dead skin squames for prolonged periods in the environment under optimal climatic conditions (5). Ryser-Degiorgis et al., (2002) documented the capture of an asymptomatic carrier Lynx in Switzerland that died from sarcoptic mange three months later. The course and clinical manifestation of the disease is related to the underlying immune status of the individual (1). Consequently, there is a low likelihood that <i>Sarcoptes scabei</i> could be translocated to the UK in the population of asymptomatic Trial Lynx, and reactivated to result in clinical disease as a result of stress induced immunosuppression. Given the prolonged incubation period and potential for environmental contamination from untreated, diseased Lynx, the overall likelihood of release is moderate.

As Sarcoptic scabei is present in both the source and release environments, the potential exists for density **Exposure Assessment** dependent transmission from infected foxes and other prey species in the destination environment to the Lynx population and vice-versa. Sarcoptic mange can be acquired through direct contact with infected individuals or though indirect contact with the environment (1). As Sarcoptic mange in red foxes is endemic at a low prevalence level in the Kielder area (5), Lynx could acquire sarcoptic mange by preying on diseased foxes. Furthermore, despite the solitary nature of Lynx, intraspecific transmission of Sarcoptes scabei could occur between adult females and dependent kittens or between adult males and females as a result of mating. The potential for zoonotic infections is low but scientists and field veterinarians could become infected as a result of handling anaesthetized symptomatic Lynx or infected fomites during capture and health screening procedures. Overall the risk of exposure from infected founder Lynx is low, but the risk of transmission to Lynx from infected wildlife at the destination environment is moderate. Epizootics of sarcoptic mange have not been demonstrated to have long-term population level effects in stable Consequence free-ranging Lynx populations (8, 1). However, mortality related to infectious disease can threaten the viability of Assessment small translocated Lynx populations in the context of additional sources of anthropogenic mortality such as poaching (7). As Sarcoptic mange is endemic at a low prevalence level in Kielder Forest (5), the small number of translocated Lynx pose no significant threat in terms of disease transmission to wildlife, livestock or domestic pets in the destination environment. However, the interspecific disease transmission from infected foxes to Lynx and the subsequent intraspecific transmission within the population of Trial Animals could result in significant morbidity and mortality, reducing reproductive potential and recruitment. In addition to the biological impacts of disease, the loss of a small number of adult breeding Lynx, could threaten the viability of the reintroduction programme with far reaching economic consequences such as an increased requirement for disease monitoring and management. Despite the potential for severe biological and economic consequences of sarcoptic mange in a population of

translocated Lynx, foxes only form a small part of the diet of Lynx and the incidence of Sarcoptic mange in foxes in

Kielder is low. Therefore, the overall likelihood of consequences as a result of *Sarcoptes scabei* is moderate.

Overall Risk	Moderate
Estimation	
Risk Management	To mitigate the risk of <i>Sarcoptes scabei</i> , all founder individuals will be tested for the ectoparasite (Sarcoptes Antibody ELISA and skin scrapes) prior to leaving the source environment. Following testing, all individuals will be treated with ivermectin injectable solution which is the treatment of choice for sarcoptic mange.
	Surveillance of Lynx in the soft release enclosure and free-ranging Lynx and red foxes in the destination environment, will determine the spatial and temporal prevalence of sarcoptic mange in those species. The identification of individual diseased Lynx will enable the capture, treatment and rehabilitation of those individuals.

- 1. Pence, D.B. and Ueckermann, E. (2002) Sarcoptic manage in wildlife. Revue scientifique et technique (International Office of Epizootics), 21(2), pp.385-398.
- 2. Bornstein, S., Mörner, T. and Samuel, W.M. (2001) Sarcoptes scabiei and sarcoptic mange. Parasitic Diseases of Wild Mammals, Second Edition, pp.107-119.
- 3. Mörner, T. (1992) Sarcoptic mange in Swedish wildlife. Revue scientifique et technique (International Office of Epizootics), 11(4), pp.1115-1121.
- 4. Wildlife disease monitoring in Sweden (2014) National Veterinary Institute, SVA, Uppsala, Sweden. SVA's report series 33, ISSN 1654-7098. Available at: http://www.sva.se/globalassets/redesign2011/pdf/om sva/publikationer/vilda-djur/wildlife-disease-monitor-2014.pdf. (Accessed: 18.12.16)
- 5. Soulsbury, C.D., Iossa, G., Baker, P.J., Cole, N.C., Funk, S.M. and Harris, S. (2007) The impact of sarcoptic mange Sarcoptes scabiei on the British fox Vulpes vulpes population. *Mammal Review*, *37*(4), pp.278-296.
- 6. Mörner, T. and Christensson, D. (1984) Experimental infection of red foxes (Vulpes vulpes) with Sarcoptes scabiei var. vulpes. *Veterinary Parasitology*, *15*(2), pp.159-164.
- 7. Ryser-Degiorgis, M.P., Ryser, A., Bacciarini, L.N., Angst, C., Gottstein, B., Janovsky, M. and Breitenmoser, U. (2002) Notoedric and sarcoptic mange in free-ranging lynx from Switzerland. *Journal of Wildlife Diseases*, *38*(1), pp.228-232.
- 8. Ryser-Degiorgis, M.P. (2009) Causes of mortality and diseases of Eurasian lynx (Lynx lynx). *Iberian lynx ex-situ conservation: An interdisciplinary approach*, pp.274-289.

Mycobacterium bovis	Hazard categories: Destination
Justification for Hazard Status	Mycobacterium bovis (M. bovis) is a Gram positive, acid-fast bacterium in the Mycobacterium tuberculosis complex of the family Mycobacteriaceae. It has a global distribution, a wide host range, and can persist for several months in the environment (1). Infection is acquired by inhalation, ingestion or via breaks in the skin, and bacilli can be shed in respiratory secretions, faeces, urine, pus, milk, vaginal secretions and semen (1 and 2). M. bovis is the cause of bovine tuberculosis (bTB), and as such is a major cause of economic loss worldwide. Eradication of bTB in many countries has been hindered by the fact that M. bovis persists within wildlife reservoir hosts – in the UK the badger (Meles meles) is widely believed to be the primary reservoir (or maintenance) host. M. bovis has been notifiable in the UK since 2006 (3). Although reports of TB in wild carnivores are rare (4), the Iberian lynx (Lynx pardinus) is known to be susceptible to M. bovis infection (5). Infection has been reported in two free-living (4 and 6) and two captive Iberian lynx (7). Although there are no reports of M. bovis infection in free-living Eurasian Lynx (Lynx lynx), an outbreak of tuberculosis (TB) due to M. bovis that affected two captive Lynx has been reported (8). Sweden is currently classified as Officially TB Free (OTF), a recognition of the low and stable incidence of bTB in Swedish herds (15). Furthermore, there are no reports of M. bovis in free-living Swedish Lynx (9, 10 and 11). M. bovis therefore constitutes a destination hazard, and a release assessment is not required
Risk Assessment	
Exposure Assessment	Although Scotland has been OTF since September 2009, England and Wales have never achieved the same status. In the UK, bTB is concentrated in the west and south-west of England, as well as in Wales (3). In low risk areas (the north, east and south-east of England) the incidence of bTB is very low and stable, and the majority of breakdowns can be linked to movements of undetected infected cattle from other areas of the UK (12). Scotland's herd incidence is very low and stable and is also largely driven by introductions of infected cattle (12). The current bTB surveillance and control scheme has been successful at preventing the establishment of disease in many counties in the north and east of England, areas that are thought to not yet have a significant reservoir of infection in wildlife (13). There has never been a systematic approach to bTB surveillance in non-bovine species in the UK, however, and the disease is believed to be spreading north and east, so there is no room for complacency (3).

While badgers are known to be the primary maintenance host for *M. bovis* in the UK (1) several species of deer (as well as foxes, hares and rodents) have been identified as spill-over hosts (1). Deer are highly susceptible to *M. bovis* (2), and are believed to be able to act as maintenance hosts in certain situations (such as when population densities are very high) (3). Disease in deer tends to be subacute or chronic in nature (1), and a respiratory route of infection seems most important (2). MAFF investigations that ran from the 1970s to the 1990s found a 0.9% prevalence of *M. bovis* in UK roe deer (*Capreolus capreolus*) (2). MAFF's collection of carcases was targeted around areas of high TB incidence in cattle, so does not represent disease prevalence across all UK wildlife populations – disease prevalence in areas of low bTB incidence may be lower still.

Lynx will target roe deer in poor condition, because they are easier to catch (5) and so exposure to *M. bovis* through their primary prey species remains possible, even if disease prevalence in roe deer is low. Disease exposure through predation on other species appears relatively unlikely. There are no published cases of *M. bovis* infection in wild rabbits (*Oryctolagus cuniculus*), European hares (*Lepus europaeus*) or mountain hares (*Lepus timidus*) in the UK (2). A very low to zero *M. bovis* prevalence in voles, mice, shrews, rats and squirrels has also been reported (2). Furthermore, there are no references to confirmed *M. bovis* infections in feral goats or feral wild boar (*Sus scrofus*) within the UK – despite the prevalence of infection in wild boar populations elsewhere in Europe (2). TB in sheep is uncommon (3). MAFF reported a 1.2% prevalence of TB in red foxes (*Vulpes vulpes*) (2), so exposure to *M. bovis* through predation on foxes appears possible. Fox carcasses were only collected from areas of endemic bTB, however – suggesting that the prevalence in the Study Area may be lower (2).

It has been suggested that *M. bovis* spreads from infected badgers to domestic cats following interspecific aggression (14), and it is possible that Lynx could acquire *M. bovis* infection in the same way. Although cats (and carnivores in general) usually acquire *M. bovis* infection through the ingestion of infected food (4) they can also become infected by the respiratory route, or percutaneously via bites and scratches (1). A review of mycobacterial infections in domestic cats in the UK found *M. bovis*, *M. microti* and *M. avium* to have a discrete, almost entirely non-overlapping geographical distribution (14). *M. bovis* infections were concentrated in areas of endemic bTB. There were no *M. bovis* infections reported in northern England or southern Scotland – this area being dominated by *M. microti* infections. It may well be that mycobacterial infections in Trial Animals would tend to follow a similar pattern.

Exposure risk: low

Consequence Assessment	Although individual Trial Animals are likely to be at some risk of exposure to <i>M.</i> bovis, intra-specific disease transmission seems quite unlikely, given the Lynx's solitary nature (4). Furthermore, because only six Trial Animals will be released, the potential for inter-specific disease spread through environmental contamination is low. Because cats are only spill-over (or even dead-end) hosts, however, there is no evolutionary pressure on <i>M. bovis</i> to adapt to lower virulence when infecting them (15) (see Footnotes 8 and 9). <i>M. bovis</i> is therefore capable of causing severe disease in felids and it has been suggested that <i>M. bovis</i> could be highly detrimental to small and fragmented populations of Iberian lynx (4). A small population of Trial Animals could also be vulnerable. Consequence risk: moderate
Overall Risk Estimation	Low
Risk Management	Because the overall risk posed by M. bovis is low, few mitigation measures will be required. When translocated Lynx or their offspring die, however, they will receive a prompt and thorough post mortem examination. Suspicious lesions and enlarged lymph nodes will be biopsied. The biopsies will be sectioned – one sample will be sent for histology and the other frozen pending the histology result. If the histology results are suggestive of <i>M. bovis</i> infection the frozen sample will be required for further clarification of disease status.

- 1. The Center for Food Security and Public Health/Institute for International Cooperation in Animal Biologics (2009). Bovine Tuberculosis. Pages 1-6
- 2. Delahay, R. J., De Leeuw, A. N. S., Barlow, A. M., Clifton-Hadley, R. S. and Cheeseman, C. L. (2002). The status of *Mycobacterium bovis* infection in UK wild mammals: a review. The Veterinary Journal 164, 90-105
- 3. Broughan, J. M., Downs, S. H., Crawshaw, T. R., Upton, P. A., Brewer, J. and Clifton-Hadley, R. S. (2013). *Mycobacterium bovis* infections in domesticated non-bovine mammalian species. Part 1: Review of epidemiology and laboratory submissions in Great Britain 2004-2010. Vet. J. 198(2), 339-345
- 4. Briones, V., De Juan, L., Sánchez, C., Vela, A.-I., Galka, M., Montero, N., Goyache, J., Aranaz, A., Mateos, A. and Domínguez, L. (2000). Bovine tuberculosis and the endangered Iberian Lynx. Emerging Infectious Diseases 6(2), 189-191
- 5. Gortázar, C., Delahay, R. J., McDonald, R. A., Boadella, M., Wilson, G., Gavier-Widen, D. and Acevedo, P. (2012). The status of tuberculosis in European wild mammals. Mammal Rev. 42(3), 193-206
- 6. Millán, J., Candela, M. G., Palomares, F., Cubero, M. J., Rodríguez, A., Barral, M., De La Fuente, J., Almería, S. and León-Vizcaíno, L. (2009). Disease threats to the endangered Iberian Lynx (*Lynx pardinus*). The Veterinary Journal 182(1), 114-124
- 7. Martínez, F., Manteca, X. and Pastor, J. (2013). Retrospective study of morbidity and mortality of captive Ibrian Lynx (*Lynx pardinus*) in the ex situ conservation programme (2004-June 2010). Journal of Zoo and Wildlife Medicine 44(4), 845-852

- 8. Schmidbauer, S. M., Wohlsein, P., Kirpal, G., Beineke, A., Müller, G., Müller, H., Moser, I. and Baumgartner, W. (2007). Outbreak of *M. bovis* infection in a wild animal park. Vet. Rec. 161(9), 304-307
- 9. Ryser-Degiorgis, M.-P., Hofmann-Lehmann, R., Leutenegger, C. M., Hård af Segerstad, C., Mörner, T., Mattsson, R. and Lutz, H. (2005). Epizootiologic investigations of selected infectious disease agents in free-ranging Eurasian Lynx from Sweden. Journal of Wildlife Diseases 41(1), 58-66
- 10. Andrén, H., Samelius, G., Segerström, P., Sköld, K., Rauset, G.-R. and Persson, J. (2010). Mortality and poaching of lynx in Sweden. A report to the World Wide Fund for Nature (Sweden)
- 11. Bröjer, C., Hestvik, G., Neimanis, A., Malmsten, J., Mörner, T., Uhlhorn, H. and Ågren, E. (2014). Wildlife disease monitoring in Sweden, 2014. National Veterinary Institute, SVA, Uppsala, Sweden. SVA report series 33, ISSN 1654-7098
- 12. Quarterly Publication of National Statistics on the Incidence and Prevalence of Tuberculosis in Cattle in Great Britain. DEFRA. Released 14/12/16
- 13. The Strategy for achieving Officially Bovine Tuberculosis Free status for England. DEFRA. April 2014
- 14. Gunn-Moore, D. A., McFarland, S. E., Brewer, J. I, Crawshaw, T. R., Clifton-Hadley, R. S., Kovalik, M. and Shaw, D. J. (2011). Mycobacterial disease in cats in Great Britain: 1) Culture results, geographical distribution and clinical presentation of 339 cases. Journal of Feline Medicine and Surgery 13, 934-944
- 15. Hardstaff, J. L., Marion, G., Hutchings, M. R. and White, P. C. L. (2014). Evaluating the tuberculosis hazard posed to cattle from wildlife across Europe. Research in Veterinary Science 97, S86-S93

Francisella tularensis is a bacterial pathogen which causes tularaemia, a disease with potentially severe clinical	
one of only two species in the Francisellaceae family	
and is associated with typical Lynx prey species such	
t has been suggested that the Lynx could be	
orted cases. Tularaemia is a zoonotic disease with the	
ional level (5). <i>F. tularensis</i> is not transmitted from	
ting (3). Because the UK is free of <i>F. tularensis</i> (1) it is	
araemia is a disease of complex epidemiology that is	
i). Many aspects of the disease remain poorly	
5).	
en has the second highest average incidence rate in	
se within the country (5) (see Footnotes 3 and 4).	
efore could become infected with <i>F. tularensis</i> (6) (see	
993 and 1999, however, found no evidence of	
o F. tularensis have been found in Bobcat (Lynx rufus)	
occasionally reported in the domestic cat (Felis catus)	
he disease (13).	
ents and lagomorphs (1) (see Footnotes 7 and 8). It is	
spread of <i>F. tularensis</i> within the environment (3 and	
F. tularensis prevalence of 2.1% has been	
ents pose a low to moderate risk of associated	
ensis or of clinical tularaemia in the Lynx, and despite	
ceptible to tularaemia, the risk of Trial Animals	
a wide array of arthropod vectors (17). Strong	
ctors of <i>F. tularensis</i> (5) and may also be significant	

reservoirs of infection (17 and 18) (see Footnote 9). The prevalence of *F. tularensis* in European tick populations is generally between 0 and 3%, but can be higher (20). The tick's relatively long life span (c. 2 years) permits it to transmit infection several times over relatively long periods of time (17).

Transmission of the organism by mosquitoes has also been demonstrated (3) (see Footnote 10). Mosquitoes are believed to be mechanical rather than biological vectors of disease, transmitting the organism transiently on contaminated mouthparts via interrupted feeding (17 and 18). It has been suggested that mosquitoes play a greater role in the epidemiology of tularaemia in northern Europe, where they are relatively abundant and ticks relatively rare (22). Clinical experience and epidemiological data support the role of mosquitoes as vectors of *F. tularensis* in Sweden (23 and 24) (see Footnote 11).

Tabanid flies have also been associated with tularaemia outbreaks (28) and several species have been shown to carry *F. tularensis* (17). They are believed to act as mechanical rather than biological vectors, transmitting infection transiently via contaminated mouthparts during interrupted feeding (see Footnote 12).

F. tularensis has been found in fleas – but their role in the epidemiology of tularaemia is currently unclear (13). Swedish Lynx are undoubtedly affected by various arthropod species suspected of involvement in the epidemiology of tularaemia, but the risk of Lynx becoming infected as a result appears to be low. Arthropods such as ticks, tabanids, mosquitoes and fleas (upon translocated Lynx or in the transport module) all pose a moderate to high associated invasion risk in the absence of risk mitigation, however.

The incidence of human tularaemia across Europe varies by season – there are more cases in summer and autumn (5 – Figure 2). Capturing the Trial Animals during these months is likely to increase the risk of releasing *F. tularensis* into the Study Area. Taking Trial Animals from a hotspot of endemic disease (or a region currently suffering from an epidemic) would also increase the risk.

Overall risk of release (without risk management): MODERATE.

Exposure Assessment

Infected lynx could become parasitized by blood feeding arthropods vectors (ticks, mosquitoes and tabanids) within the Study Area. They could also potentially excrete *F. tularensis* in urine or faeces (this hasn't been reported in cats, but can't be ruled out). Risk of exposure via infected Lynx: moderate.

Infected stowaway rodents could excrete *F. tularensis* in urine (14) or faeces (15). Aquatic or semi-aquatic rodents could infect water courses (see Footnote 13). Infected rodents could become parasitized by blood feeding arthropod vectors (ticks, mosquitoes and tabanids). They could also fall prey to carnivores such as foxes, domestic cats and Lynx. Rodents could also spread infection by fighting with conspecifics. Risk of exposure via infected rodents: high.

	Infected ticks can transmit infection during a blood meal. They are biological vectors to which <i>F. tularensis</i> is well adapted. The prevalence of the organism in Swedish ticks can be high and transstadial transmission occurs (17). Lynx could play host to tick larvae and/or nymphs, which could go on to parasitize a variety of animal species (including smaller species such as rodents) as adult ticks. Risk of exposure via ticks: high. Infected mosquitoes can transmit infection mechanically during interrupted feeding (21), potentially infecting many individuals. Although mosquitoes can feed repeatedly, their lifespan is short (typically a few weeks). Transovarial transmission hasn't been shown (so the infection dies with the mosquito) (17). Risk of exposure via infected mosquitoes: moderate. Infected tabanids are mechanical vectors capable of causing tularaemia outbreaks via interrupted feeding (28). They have been shown, however, to be capable of transmitting disease for only four days after initial infection (29). Risk of exposure via infected tabanids: moderate. Fleas can carry <i>F. tularensis</i> infection (13) and Lynx may play host to fleas. Although it seems unlikely, we can't exclude the possibility of fleas spreading the organism to the Study Area. A comprehensive exposure assessment should also consider the risk of exposure to humans. Zoonotic transmission could occur through aerosols, ingestion of contaminated food/water and through direct contact with infected pets, livestock, wildlife and arthropods. Risk of human exposure: moderate. Overall exposure risk: MODERATE.
Consequence Assessment	If susceptible animals or humans in the Study Area are exposed to <i>F. tularensis</i> the likelihood of consequences is high, because the organism is highly infectious. <i>F. tularensis</i> is notifiable to the OIE when first diagnosed in animals, and notifiable to Public Health England if diagnosed in humans. Doctors are likely to be slow to diagnose the condition, due to lack of familiarity and a long list of differentials. A tularaemia outbreak in the UK would necessitate a huge mobilisation of resources and the involvement of a wide variety of personnel (vets, biologists, epidemiologists, government agency staff etc). A "runaway" epidemic would not occur, however, because human to human transmission of <i>F. tularensis</i> does not occur (3 and 1). It also seems very unlikely that a significant outbreak would occur in the absence of a substantial pre-existing environmental reservoir of infection. Overall consequence severity: MODERATE.
Risk Estimation	Given a moderate risk of release and a moderate risk of exposure, we can conclude that the overall likelihood of entry and exposure is moderate . The consequences of entry and exposure are also moderate . Use of the risk estimation matrix suggests that the overall risk posed by <i>F. tularensis</i> (without risk management) is MODERATE .

Risk Management	We will avoid trapping lynx in areas where tularaemia is endemic (e.g. Norbotten, Ljusdal) or emergent (e.g. Örebro), and we will consider the possibility of trapping Lynx in areas where the Flood Water Mosquito (<i>Aedes sticticus</i>) doesn't occur (i.e. areas of the north where tularaemia isn't endemic). We will, if possible, avoid trapping Lynx during seasons of high tularaemia incidence (the summer and autumn) – although this decision will also be affected by other factors related to Lynx biology and to logistics. It is also worth noting that Swedish Lynx are unlikely to have encountered the organism, whatever time of year they are trapped. We will liaise with local public health and veterinary health authorities in Sweden, so that we are fully aware of recent local tularaemia outbreaks in either humans or animals. All captured Lynx will be anaesthetised and carefully checked for symptoms of tularaemia (as per 9 and 12). All Lynx will be tested for antibodies to <i>F. tularensis</i> (see Footnotes 14 and 15). We also hope to be able to perform PCR testing (on blood) to look for the organism itself. We will test a proportion of the ticks that we find on captured Lynx for <i>F. tularensis</i> . All translocated Lynx are to be treated for ticks with fipronil (which will also kill fleas). We will take measures to ensure that no associated invasion (of ticks, mosquitoes, tabanids, fleas or rodents) occurs. Any Lynx fatalities occurring after translocation will receive a full and prompt post-mortem examination (including fluorescent antibody testing of tissue samples, where appropriate). The implementation of these mitigation measures would reduce the risk of release and exposure in the Study Area to very low levels. Use of the risk estimation matrix suggests that the overall post-mitigation risk posed by <i>Francisella tularensis</i> is LOW.
Footnotes	 There are four subspecies of <i>F. tularensis</i>, but the only subspecies found in Europe is <i>F. tularensis</i> holoarctica – also known as <i>F. tularensis</i> type B. It causes a milder disease than <i>F. tularensis</i> type A (7) and is generally non-lethal in humans (1). Because doctors in the UK are generally unfamiliar with the condition, and because the list of differentials is long (3), diagnosis of tularaemia in human patients in the event of an outbreak in the UK is unlikely to be swift. Sweden was responsible for 25% of all reported European tularaemia cases between 1992 and 2012 (5). Some authors have stated that <i>F. tularensis</i> is only endemic in the north of the country (1). Between 1992 and 1998, 80% of reported human tularaemia cases in Sweden occurred in the north of the country (8). In Norrbotten (in the far north of the country) in 2012 there were 82 cases per 100,000 people (5). However the pattern is not static – in the 2000s the disease emerged in areas of central Sweden too (4).

- 5. Cats are believed to become infected by *F. tularensis* via 1) direct contact with or ingestion of infected animal tissue, 2) ingestion of contaminated water, 3) arthropod bites or 4) inhalation of aerosols (9).
- 6. This may have been because most of the Lynx carcasses were tested during the winter, when tularaemia in Scandinavian hare populations is at a low level (10). However because *F. tularensis* antibody titres in Lynx are likely to persist for several years (11), it is more likely that the Lynx had genuinely never been exposed to the organism.
- 7. Voles, mice, lemmings and hares are considered to be highly susceptible to infection (3). Infected voles have been shown to shed the organism via both urine (14) and faeces (15), and outbreaks of tularaemia in humans often follow outbreaks in rodents (3). The shedding of the organism by chronically infected hares has not been shown, but is certainly possible (1).
- 8. There is no evidence, however, that mammals constitute a major reservoir of infection (3). Although highly susceptible to infection, rodents and lagomorphs do not seem capable of harbouring the bacteria between outbreaks (1). This may be because they rarely survive the infection (1) and therefore represent accidental rather than reservoir hosts of the bacterium (7).
- 9. *F. tularensis* is known to disseminate to and replicate within the tick's salivary glands, thus entering the saliva (19). They are known to be consequently capable of transmitting an infection during a blood meal (19) and transstadial transmission is known to occur (17). Transovarial transmission has not been demonstrated, however (17).
- 10. It has been demonstrated that multiple mosquito species (including *Aedes* species) can transmit disease to mice (21).
- 11. A high prevalence of the organism has been demonstrated in Swedish mosquitoes (25) and a correlation between mosquito abundance and human tularaemia cases in forested parts of the country has been reported (26). Aedes sticticus (the Flood Water Mosquito) and Aedes cinereus are the species of greatest concern in terms of the spread of tularaemia within Sweden (27). A. cinereus is found across the whole country, whereas A. sticticus is confined to the south (27). The distribution of tularaemia in eastern Europe and Sweden is related to natural water (1) and it has been suggested that mosquitoes may be responsible for the spread of F. tularensis from a water reservoir to humans (5). The larvae of Flood Water Mosquitoes (A. sticticus) prey on aquatic protozoa, and may well be exposed to F. tularensis this way (4).
- 12. The organism is thought to neither multiply nor survive long term within tabanids (17). The deer fly (*Chrysops discalis*) is able to transmit *F. tularensis* to animals for only four days following infection (29).

- 13. *F. tularensis* is thought to be able to survive for long periods in aquatic protozoa, which could in turn lead to infection becoming acquired by mosquito larvae. The organism is known to be transmitted transstadially in mosquitoes, so infected larvae could soon become infected adult mosquitoes with the ability to act as mechanical vectors of disease.
- 14. It should be noted that studies have found only 1 of 3 domestic cats with tularaemia to be seropositive (12).
- 15. Ideally paired sera will be taken in order to identify a rising antibody titre that could reflect recent exposure to *F. tularensis* or the current incubation of disease.
- 1. Tärnvik, A. and Berglund, L. (2003). Tularaemia. Eur. Respir. J. 21, 361-373
- 2. Collins, F. M. (1996). Pasteurella, Yersinia and Francisella. Medical Microbiology, 4th edition. Chapter 9
- 3. WHO Guidelines on Tularaemia (2007). ISBN 978 92 4 154737 6
- 4. Broman, T., Thelaus, J., Andersson, A.-C., Bäckman, S., Wikström, P., Larsson, E., Granberg, M., Karlsson, L., Bäck, E., Eliasson, H., Mattsson, R., Sjöstedt, A. and Forsman, M. (2011). Molecular detection of persistent *Francisella tularensis* subspecies holarctica in natural waters. Int. J. Microbiol. 851946
- 5. Hestvik, G., Warns-Petit, E., Smith, L. A., Fox, N. J., Uhlhorn, H., Artois, M., Hannant, D., Hutchings, M. R., Mattsson, R., Yon, L. and Gavier-Widen, D. (2015). The status of tularaemia in Europe in a one-health context: a review. Epidemiol. Infect. 143, 2137-2160
- 6. Ryser-Degiorgis, M.-P., Hofmann-Lehmann, R., Leutenegger, C. M., Hård af Segerstad, C., Mörner, T., Mattsson, R. and Lutz, H. (2005). Epizootiologic investigations of selected infectious disease agents in free-ranging Eurasian Lynx from Sweden. Journal of Wildlife Diseases 41(1), 58-66
- 7. Mörner, T. (1992). The ecology of tularaemia. Revue Scientifique et Technique (International Office of Epizootics) 11(4), 1123-1130
- 8. Ekdahl, K. and Twisselmann, B. (2001). Epidemics of tularaemia in Sweden and Finland. Eurosurveillance 5, 1825
- 9. Woods, J. P., Panciera, R. J., Morton, R. J. and Lehenbauer, T. W. (1998). Feline tularaemia. Compendium on Continuing Education for the Practicing Veterinarian 20. 442-457
- 10. Mörner, T. (1988). Infections with Francisella tularensis biovar palaearctica in hares (Lepus timidus, Lepus europaeus) from Sweden. J. Wildl. Dis. 24(3), 422-433
- 11. Ericsson, M., Sandström, G., Sjöstedt, A. and Tärnvik, A. (1994). A persistence of cell-mediated immunity and decline of humoral immunity to the intracellular bacterium *Francisella tularensis* 25 years after natural infection. J. Infect. Did. 170, 11-114
- 12. Baldwin, C. J., Paciera, R. J., Morton, R. J., Cowell, A. K. and Waurzyniak, B. J. (1991). Acute tularaemia in three domestic cats. Journal of the American Medical Association 199(11), 1602-1605
- 13. Olsufiev, N. G. and Dunayeva, T. N. (1970). Natural focality, epidemiology and prophylaxis of tularaemia. Medicina 9, 11-18
- 14. Bell, J. F. and Stewart, S. J. (1975). Chronic shedding tularaemia nephritis in rodents: possible relation to occurrence of *Francisella tularensis* in lotic waters. J Wildl. Dis. 11(3), 4231-430
- 15. Dahlstrand, S., Ringertz, O. and Zetterberg, B. (1971). Airborne tularaemia in Sweden. Scandinavian Journal of Infectious Diseases 3, 7-16
- 16. Olsufiev, N. G. (1977). Results and perspectives of the study of natural foci of tularaemia in USSR. Meditsinskaia parazitologiia parazitarnye bolezni 46, 273-282
- 17. Petersen, J. M., Mead, P. S. and Schriefer, M. E. (2009). Francisella tularensis: an arthropod-borne pathogen. Vet. Res. 40(2), 07

- 18. Akimana, C. and Kwaik, Y. A. (2011). *Francisella*-arthropod vector interaction and its role in patho-adaptation to infect mammals. Frontiers in Microbiology Vol. 2, Article 34, pages 1-14
- 19. Reif, K. E., Palmer, G. H., Ueti, M. W., Scoles, G. A., Margolis, J. J., Monack, D. M. and Noh, S. M. (2011). *Dermacentor andersoni* transmission of *Francisella tularensis* subsp. *Novicida* reflects bacterial colonization, dissemination and replication coordinated with tick feeding. Infect. Immun. 79(12), 4941-4946
- 20. Hubálek, Z. and Halouzka, J. (1997). Mosquitoes, in contrast to ticks, do not carry *Francisella tularensis* in a natural focus of tularaemia in the Czech Republic. J. Med. Entomol. 34(6), 660-663
- 21. Philip, C. B. and Parker, R. R. (1932). Experimental transmission of tularaemia by mosquitoes. Public Health Rep. 47, 2077-2088
- 22. Keim, P., Johansson, A. and Wagner, D. M. (2007). Molecular epidemiology, evolution and ecology of *Francisella*. Annals of the New York Academy of Sciences 1105, 30-66
- 23. Christensen, B. (1984). An outbreak of tularaemia in the northern part of central Sweden. Scandinavian Journal of Infectious Diseases 16, 285-290
- 24. Eliasson, H. et al. (2002). The 2000 tularaemia outbreak: a case-control study of risk factors in disease-endemic and emergent areas, Sweden. Emerging Infectious Diseases 8, 956-960
- 25. Lundström, J. O., Andersson, A.-C., Bäckman, S., Schäfer, M. L., Forsman, M. and Thelaus, J. (2011). Transstadial transmission of *Francisella tularensis holarctica* in mosquitoes, Sweden. Emerg. Dis. 179%0, 794-799
- 26. Rydén, P., Björk, R., Schäfer, M. L., Lundström, J. O., Petersén, B., Lindblom, A., Forsman, M., Sjöstedt, A. and Johansson, A. (2012). Outbreaks of tularaemia in a boreal forest region depends on mosquito prevalence. J. Infect. Dis. 205(2), 297-304
- 27. Lundström, J. O., Schäfer, M. L., Hesson, J. C., Blomgren, E., Lindström, A., Wahlqvist, P., Halling, A., Hagelin, A., Ahlm, C., Evander, M., Broman, T., Forsman, M. and Persson Vinnersten, T. Z. (2013). The geographic distribution of mosquito species in Sweden. Journal of the European Mosquito Control Association 31, 21-35
- 28. Klock, L. E., Olsen, P. F. and Fukushima, T. (1973). Tularaemia epidemic associated with the deerfly. Jornal of the American Medical Association 226, 149-152
- 29. Francis, E. and Mayne, B. (1921). Experimental transmission of tularaemia by flies of the species Chrysops discalis. Public Health Rep. 36, 1738-1746

F) Generic Ticks	Hazard Categories (Source/Destination/Zoonoses)
	The haematophagous, parasitic activities of ticks are generally associated with low levels of morbidity in Lynx. However, the hazard status of ticks relates to their role as important vectors in the transmission of disease and the existence of novel species in the source and destination environments. In Northern Europe the majority of tick species belong to the family Ixodidae with a limited number belonging to the family Argasidae (1). <i>Ixodes ricinus</i> , the sheep tick, is the most abundant and widely distributed hard tick species in Sweden and in the UK (1).
	Despite significant overlap in the tick fauna of the UK and Sweden, the British Isles are more species rich due to a conducive climate and more southerly latitude (1). Most species in the family ixodidae occur in both the source and destination environment although some species such as <i>Dermacentor reticulatus</i> and <i>Hyalomma marginatum</i> are only present in the UK (2, 1). Climate change scenarios are predicted to alter the abundance and distribution of tick species in Northern Europe, due to increased winter warming and associated tick survival (3). Furthermore, the movement of livestock and domestic pets could enable southern European species to extend their northern range and colonise countries such as Great Britain (3). Therefore, there is a potential risk that the translocation of Lynx to a destination environment at a more southerly latitude could expose them to novel tick species and increased tick densities.
	Ixodid ticks constitute a vector for the transmission of many diseases including <i>Anaplasma</i> , <i>Babesia</i> , <i>Borrelia and Flavivirrus</i> species. However, the majority of tick borne diseases occur as the same subspecies in the source and destination environments and are not novel to the Lynx. Only tick-borne encephalitis virus and tularaemia constitute zoonotic source hazards and comprehensive risk assessments relating to these diseases can be found in Appendix 1 of this report. Given the mitigation measures to avoid the entry of novel tick species and associated diseases from the source environment, the overall risk to Lynx from ticks in the destination environment is low. Despite the likelihood of exposing some founder Lynx from more northerly parts of Sweden to higher tick densities in the destination environment, the Lynx will only constitute an accidental host for tick-borne diseases in the UK. Therefore, it is unlikely that the Lynx will be impacted at a population level by tick-borne diseases in the UK and furthermore will not act as a reservoir for the transmission of zoonotic tick-borne infections in the release environment such as louping ill.

- 1. Jaenson, T.G., TäLleklint, L., Lundqvist, L., Olsen, B., Chirico, J. and Mejlon, H., 1994. Geographical distribution, host associations, and vector roles of ticks (Acari: Ixodidae, Argasidae) in Sweden. *Journal of Medical Entomology*, *31*(2), pp.240-256.
- 2. European Centre for Disease Prevention and Control (ECDC) (2016) Tick maps. Available at: http://ecdc.europa.eu/en/healthtopics/vector
- 3. Gray, J.S., Dautel, H., Estrada-Peña, A., Kahl, O. and Lindgren, E., 2009. Effects of climate change on ticks and tick-borne diseases in Europe. *Interdisciplinary perspectives on infectious diseases*, 2009.

Appendix 2.
Individual Disease Risk Assessments: Welfare Hazards

A) Illegal Persecution (Shooting/Trapping /Poisoning)	Hazard Categories (Destination/Population)
Justification for Hazard Status	Non-infectious causes of mortality such as illegal shooting, trapping or poisoning constitute the greatest threat to Lynx populations across Europe (1). In contrast the overall prevalence of Infections in Lynx is low due to their solitary nature, and epizootics of disease are rarely reported (2).
	Between 1994 and 2002 the cause of mortality was documented for 245 radio-collared Lynx at five study sites in Sweden and Norway (3). Poaching accounted for 46% of fatalities in adult Lynx due to conflict with reindeer (<i>Rangifer tarandus</i>) herders in Northern Scandinavia and roe deer (<i>Capreolus capreolus</i>) hunters in Southern regions (3). A study conducted between 1987 and 1999 in the Swiss Alps and Jura mountains, established that 72% of deceased Lynx (57/72) submitted to the Institute of Animal Pathology, Institute of Bern, had died of anthropogenic causes (4). Of those, 15 had died in road traffic accidents, 8 had been poached and two had died as a result being trapped. Death due to infectious disease only accounted for 18% (13/72) of the overall mortality (4).
	Anthropogenic mortality does not generally threaten the viability of free-ranging autochthonous Lynx populations, however illegal persecution is often additive to other forms of mortality such as infectious disease (3), and could have population level impacts on a small reintroduced Lynx population in the UK.
Risk Assessment	, , , , , , , , , , , , , , , , , , ,
Exposure Assessment	Data sets relating to wildlife crime in the UK are held by both government and non-government agencies. The annual Birdcrime report published by the The Royal Society for the Protection of Birds (RSPB) is the most comprehensive source of raptor persecution incident data for British birds in space and time (5). Extrapolating the risk to Lynx from the Birdcrime report is justified on the basis that both species could be perceived as threats to the interests of rural stakeholders. Data from the Birdcrime (2014) report document a decrease in the reported incidence of 'Shooting and destruction of birds of prey' from 284 in 2009 to 179 in 2014. Furthermore, similar reductions were reported for 'Poisoning and use of poison baits' from 158 to 72 in 2009 and 2014 respectively (5). Since 2012 the Scottish

Government Environment and Forestry Directorate has published annual reports on wildlife crime in Scotland (6). The 2015 report records a decrease of 20% in wildlife crime reported to Police Scotland from 355 incidents in 2010-11 to 284 in 2014-15 (6).

The ongoing and illegal use of firearms and poisons could represent a threat to the Trial Animals in the UK. In contrast to shooting and trapping, which requires a sustained effort by criminals, poisoning can have a population level impact with only minimal effort and poison baits continue to be lethal over a period of days or weeks without further effort by the poisoner. Lynx could be exposed to illegal poisons such as carbofuran as they have been documented to consume carrion if live prey is scarce through the winter months (7).

Many studies have analyzed the spatial distribution of illegal persecution incidents and have documented an association with open moorland areas managed for high value driven red grouse shooting (5). The reintroduced Lynx are unlikely to use open moorland areas where they might encounter poison baits and the Kielder forest is not associated with high value field sports.

Overall the likelihood of exposure to illegal persecution is moderate on the basis of reductions in the incidence of wildlife crime in the UK, a low chance of exposure to grouse moors and a moderate likelihood of conflict with rural stakeholders.

Consequence Assessment

The death of even a small number of Lynx from the founder population could have severe biological and economic impacts on the viability of the Trial. The loss of key breeding individuals could reduce reproductive potential and recruitment. Furthermore, due to the indiscriminate nature of poison baits the risk of morbidity and mortality would extend to other resident species of wild carnivores/omnivores such as badgers (*Meles meles*) and to domestic pets such as dogs walked in the Kielder forest. Economic impact would derive from the need for additional surveillance of the remaining Lynx and the requirement for comprehensive post mortem examination including toxicology screening.

The severe consequences of illegal persecution are moderated by the following factors; firstly, the introduction of robust legislation to control the illegal use of firearms, poisons and traps; secondly the implementation of legislation and surveillance by the National Wildlife Crime Unit (NWCU) and finally a moderate chance of exposure. Overall the likelihood of consequences is moderate.

Overall Risk Estimation	Moderate
Risk Management	To mitigate the threats of illegal persecution the Trust has conducted an exhaustive community engagement programme within the destination environment including public meetings, farm visits and door to door work. The aim of the local consultation ("PALC") is to inform the public, address their concerns and invite them to contribute to the work of the trust. A compensation scheme will operate to mitigate any financial losses resulting from Lynx preying on sheep.
	Satellite collars will enable the Team to identify the location of each Lynx in real time, to provide an early warning system if individual Lynx are straying beyond the forest boundary into grazed pasture and moorland. In the unlikely event of a 'rogue' Lynx targeting livestock, the individual will be recaptured and maintained in captivity or translocated to suitable habitat out-with the area of capture.

- 1. Von Arx M, Breitenmoser-Würsten C, Zimmermann F, Breitenmoser U (2004) *Status and conservation of the Eurasian lynx* (Lynx lynx) *in 2001. KORA Bericht* no. 19, Muri.
- 2. Ryser-Degiorgis, M.P. (2009) Causes of mortality and diseases of Eurasian lynx (Lynx lynx). *Iberian lynx ex-situ conservation: An interdisciplinary approach*, pp.274-289.
- 3. Andrén, H., Linnell, J.D., Liberg, O., Andersen, R., Danell, A., Karlsson, J., Odden, J., Moa, P.F., Ahlqvist, P., Kvam, T. and Franzén, R. (2006) Survival rates and causes of mortality in Eurasian lynx (Lynx lynx) in multi-use landscapes. *Biological Conservation*, 131(1), pp.23-32.
- 4. Schmidt-Posthaus, H., Breitenmoser-Würsten, Ch., Posthaus, H., Bacciarini, L. N., and Breitenmoser, U. (2002) Causes of mortality in reintroduced Eurasian lynx in Switzerland. J. Wildl. Dis. 38(1): 84-92.
- 5. RSPB (2014) Birdcrime 2014. Available at: https://www.rspb.org.uk/lmages/birdcrime_2014_tcm9-410409.pdf. (Accessed: 01.01.17).
- 6. The Scottish Government (2016) Wildlife Crime in Scotland 2015 Annual Report. Available at: http://www.gov.scot/Resource/0051/00511181.pdf. (Accessed: 08.01.17).
- 7. Odden, J., Linnell, J.D. and Andersen, R. (2006) Diet of Eurasian lynx, Lynx lynx, in the boreal forest of southeastern Norway: the relative importance of livestock and hares at low roe deer density. *European Journal of Wildlife Research*, 52(4), pp.237-244.

B) Generic Stress Hazard Categories (Source/Destination/Population/Transport) Justification for Stress can impact on the health and welfare of wild felines at all stages of the translocation pathway including capture, transport, soft release and post-release monitoring. Acute stress, activated by adverse or threatening **Hazard Status** stimuli, is an adaptive survival mechanism characterized by the rapid fight or flight response and is mediated by the sympathetic nervous system and glucocorticoid release following stimulation of the hypothalamic-pituitary-adrenal axis (HPA). An acute stress response during translocation could result in a heightened state of arousal, attempts to escape and associated trauma such as nail injuries and long bone fractures. Chronic stress occurs when the state of acute stress persists for prolonged periods and constitutes a common maladaptive response encountered during reintroduction programmes. Prolonged release of corticosteroids can result in suppression of the immune system, reproductive dysfunction, weight loss due to reduced feeding behaviour and increased parasite loads. Terio et al. (2004) conducted a comparative study to quantify fecal corticoid levels and adrenal hypertrophy as indicators of chronic stress in populations of captive and free ranging cheetah (Acinonyx jubatus). Captive individuals demonstrated significantly higher base-line fecal corticoid levels and larger adrenal cortices than free ranging cheetahs. The authors concluded that this chronic stress response was likely to contribute to the higher incidence of disease and reproductive dysfunction in captive populations. Despite low levels of stress-induced pathologies in captive Lynx, comparative studies of captive and wild free ranging Canada lynx (Lynx canadensis) populations have also documented significantly higher fecal corticosteroid metabolites (FGM) in captive individuals (2). Chronic stress in captive Canada lynx could have sub-clinical impacts and account for the poor reproductive output documented in zoological collections. Fanson et al. (2013) evaluated a variety of housing and husbandry factors on adrenocortical activity in captive Canada lynx populations. The study revealed significant increases in adrenocortical activity associated with the following factors: reductions in the size of the enclosure, reductions in the number of hiding places, the social structure of lynx and movement to a new enclosure or zoological collection. Mixed sex groups demonstrated higher FGM levels than same sex groups and lynx housed alone had the lowest levels of FGM (3).

Risk Assessment	
Exposure Assessment	Founder Lynx could experience stress at all stages of the translocation pathway including the administration of anaesthetics for procedures such as the application of satellite collars and routine health monitoring. Episodes of acute stress are likely to be associated with capture, transport and anaesthesia. Exposure to chronic stress could occur in the soft release enclosures ("Release Pens") at the release environment and in the post-release period.
Consequence Assessment	Acute stress could result in traumatic injuries to limbs, nails, gums and teeth and could contribute to anaesthetic induced morbidity and mortality. Chronic stress in the Release Pens could suppress immune function and increase susceptibility to disease with concomitant negative impacts on reproduction, hunting success and survival in the post-release environment.
Overall Risk Estimation	Moderate
Risk Management	All personnel with direct involvement in the translocation process, will adopt a quiet and calm manner around the founder Lynx. Each stage of the translocation will be meticulously planned to minimize human contact, avoid habituation and reduce the likelihood of human-Lynx conflict post-release.
	To minimize acute capture related stress and trauma, only box traps with wooden panels instead of steel mesh will be used. All traps will be fitted with electronic radio-alarms or sms alarms to determine the time of capture and enable a quick response from the scientific and veterinary teams (4). To mitigate transport related stress, the design of the IATA standard transport crates should be carefully considered and particular care should be taken when loading and unloading the Lynx. Non-sedative anxiolytic drugs will be used to avoid self-trauma in individuals that display severe stress behaviour during transport. The ambient temperature during transport will be monitored and adjusted to keep the Lynx cool and avoid thermal stress.
	To avoid chronic stress at the destination environment, the scientific team will incorporate recommendations from Fanson et al. (2013) and others into the design and management of the soft-release enclosures. Adult Lynx will be housed individually to reflect the solitary nature of autochthonous populations. Enclosures will be large and spatially heterogenous to provide environmental enrichment and places where the Lynx can hide.

The trust will use best practice methods drawn from the Norwegian School of Veterinary Science (5) and the International Zoo Veterinary Group (IZVG) regarding anaesthetic procedures and protocols for Lynx.

- 1. Terio, K.A., Marker, L. and Munson, L., 2004. Evidence for chronic stress in captive but not free-ranging cheetahs (Acinonyx jubatus) based on adrenal morphology and function. *Journal of Wildlife Diseases*, 40(2), pp.259-266.
- 2. Fanson, K.V., Wielebnowski, N.C., Shenk, T.M. and Lucas, J.R., 2012. Comparative patterns of adrenal activity in captive and wild Canada lynx (Lynx canadensis). *Journal of Comparative Physiology B*, 182(1), pp.157-165.
- 3. Fanson, K.V. and Wielebnowski, N.C., 2013. Effect of housing and husbandry practices on adrenocortical activity in captive Canada lynx (Lynx canadensis). *Animal welfare*, 22(2), pp.159-165.
- 4. Odden, J., Linnell, J.D.C., Arnemo, J. M. & Berntsen, F. (2007) Refinement of research capture techniques capture of Eurasian lynx in Norway 1995–2007. NINA Minirapport 203.
- 5. Arnemo, J., Evans, A. and Fahlman, A. (2007) *Biomedical protocols for free-ranging brown bears, gray wolves, wolverines and lynx. Norwegian School of Veterinary Science*. General Technical Report, Tromsø, Norway.

Appendix 3.

Necropsy Protocol for Lynx (*Lynx lynx***)**

Lynx carcases for necropsy will be promptly relocated to:

Dept of Veterinary Pathology and Public Health The Institute of Veterinary Science University of Liverpool Leahurst Campus, Neston, CH64 7TE

If this is not possible either freezing the entire carcase or an in situ necropsy may to take place.

If an in situ necropsy occurs, then please send the below completed reports (including digital images), by email if possible, to:

Dr Julian Chantrey Veterinary Adviser to the Lynx UK Trust University of Liverpool

Email: chantrey@liv.ac.uk

Necropsy Protocol for Lynx (*Lynx lynx***)**

Necropsy performed by:	
Agency/organisation:	
Email:	Phone number:
Address:	
ANIMAL DETAILS: Captive or free-ranging:	
Identity/name:	
Transponder:	
Sex:	Weight:
Date of birth (if known) :	Age:
Where found (including GPS location if possible)	:
Environmental conditions:	
Date of death:	
Date of necropsy:	
Proven breeder:	
History (include clinical signs and circumstances	of death):

GROSS NECROPSY FINDINGS:

Fill in details of gross necropsy findings in sections below, or circle one of NAD (no abnormality detected) or NE (not examined).

It is extremely valuable if you can take digital images of any abnormalities found

General condition: (nutritional condition, physical condition, carcass fresh or decomposed etc). NAD/NE

Skin: (NB in the case of a neonate, examine the umbilical stump and surrounding tissues) NAD/NE

Musculoskeletal system: (bones, joints, muscles)

NAD/NE

Body cavities: (fat stores, abnormal fluids)

NAD/NE

Lymphoreticular system: (spleen, lymph nodes, lymphatics, thymus)

NAD/NE

Respiratory system: (nasal cavity, larynx, trachea, lungs, regional LN's. In neonates note whether lungs float or sink in formol saline)

Digestive system: (mouth, teeth, oesophagus, stomach, intestines, liver, pancreas, mesenteric lymph nodes. In the case of a neonate, note whether milk is present in the stomach) NAD/NE

Cardiovascular system: (heart, pericardium, blood vessels)

NAD/NE

Urinary system: (kidneys, ureters, urinary bladder, urethra)

NAD/NE

Reproductive system: (testes/ovaries, uterus, vagina, penis, prepuce, accessory glands, mammary

glands, placenta)

NAD/NE

Endocrine system: (adrenals, thyroid, parathyroids, pituitary)

NAD/NE

Nervous system: (brain, spinal cord, peripheral nerves)

NAD/NE

Sensory organs: (ears, eyes)

NAD/NE

LABORATORY TESTS & DIAGNOSES:

Laboratory Tests:

Give details of all specimens submitted for bacteriology, virology, parasitology, histopathology.

Please attach reports of these tests to the completed form.

Bacteriology: Report attached: Y/N

Virology: Report attached: Y/N

Parasitology: Report attached: Y/N

Histopathology: Report attached: Y/N

Other: (specify) Report attached: Y/N

Preliminary diagnosis: FINAL DIAGNOSIS:

TISSUES FOR STORAGE

In addition to specimens submitted for diagnostic pathology, the following tissues should be preserved in 10% buffered formal saline at a ratio of 1 part tissue to 10 parts formal saline. Sections should be no thicker than 1 cm. Examples of all lesions should also be included. Tissues should be accurately labelled and stored.

Tissue Area Taken (y/n)
Adrenal Entire gland with transverse cut
Brain Sliced longitudinally along midline
Heart Entire heart after opening & examining atria, ventricles and valves from each side
Intestines 3cm lengths of duodenum, jejunum, ileum,caecum & colon Open along long axis
Kidney Section of cortex, medulla & pelvis from each
Liver 2 sections from 2 lobes with capsule and gall bladder
Lung Sections from several lobes including a bronchus
Lymph nodes Cervical, anterior mediastinal, bronchial, mesenteric and lumbar with a transverse cut
Pancreas Samples from 2 areas
Peripheral nerve 3cm section of sciatic nerve
Skeletal muscle Cross-section of thigh muscles
Skin 3cm length of full thickness abdominal skin
Spleen Cross section including capsule
Spinal cord Sections from cervical, thoracic & lumbar cord
Stomach Cardia, antrum and pylorus
Testis/ovary Entire with transverse cut
Thyroid Intact including parathyroids
Urinary bladder Cross-section
Uterus Entire with longitudinal cut into lumen

Appendix 4.

Hazard Identification Spreadsheet (see supporting excel spreadsheet)