Animals (Scientific Procedures) Act 1986

Non-technical summaries for projects granted during 2014

Volume 12

Projects with a primary purpose of:

- Protection of the natural environment in the interests of the health or welfare of human beings or animals
- Preservation of Species

Project Titles and key words

1. **Trapping strategy for effective grey squirrel control**
    - Grey squirrel, population control, trapping

2. **Positive exclusion screen trials for fish**
    - Salmon, Eel, Vertical bar screen

3. **Improving models of species interactions by including individual heterogeneity**
    - Rodents, mustelids, population dynamics

4. **Investigating virulence of, amphibian host range of and the effectiveness of the amphibian host response to ranaviruses**
    - Infectious disease, conservation, amphibian, ranaviruses

5. **Chemicals and fish Regulation**
    - Toxicity testing, environmental pollutants

6. **Biological Effects of Hazards in the Environment**
    - Behaviour, cancer, neurodegeneration

7. **Diadromous & freshwater fish: assessment & ecology**
    - Fish, Stock Assessment, Ecology, Environment, Conservation.

8. **Ecology of freshwater fish**
    - Fish, non-native fish, habitat loss, eutrophication

9. **Evaluation of rodenticides in the laboratory and field**
    - Rodenticides, rodents, resistance, anticoagulants

10. **West Country salmon and sea trout migration project**
    - Salmon, sea trout, smolts

11. **The population ecology of North Sea seabirds**
    - Seabirds, climate change, diet, physiology, demogaraphy

12. **Novel rodenticide project**
    - Rodents, Rodenticides, control, toxicity

13. **Impacts and mitigation of river structures on fish**
    - Dams, hydropower, hydrodynamics, acoustics, telemetry

<table>
<tr><td>PROJECT 1</td><td colspan="3">Trapping strategy for effective grey squirrel control</td></tr>
<tr><td>Key Words (max. 5 words)</td><td colspan="3">Grey squirrel, population control, trapping</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td colspan="3">5</td></tr>
<tr><td>Purpose of the project (as in section 5C(3)</td><td colspan="3"><table>
<tr><td>Basic research</td><td></td><td>No</td></tr>
<tr><td>Translational and applied research</td><td></td><td>No</td></tr>
<tr><td>Regulatory use and routine production</td><td></td><td>No</td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td>Yes</td><td></td></tr>
<tr><td>Preservation of species</td><td>Yes</td><td></td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td></td><td>No</td></tr>
</table></td></tr>
<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td colspan="3">To develop a tightly focussed strategy for grey squirrel trapping. In order to do this we need to know how individual squirrels respond to a choice of bait hoppers, and how they interact with each other. This requires tagging to allow individuals to be recognised.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?</td><td colspan="3">Improvement of strategy for controlling grey squirrels. This is of potential value to timber growers and red squirrel conservationists, in the UK and in increasing areas of continental Europe.</td></tr>
<tr><td>What species and approximate numbers of animals do you expect to use over what period of time?</td><td colspan="3">Grey squirrel, Sciurus carolinensis

Up to 50 per annum</td></tr>
<tr><td>In the context of what you propose to do to the animals,</td><td colspan="3">No adverse effects of tagging with collars have</td></tr>
</table>

what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	been reported and are considered unlikely, but could theoretically range from mild to severe. The intention is to recapture all tagged animals following a few weeks of monitoring. Previous work suggests that this is very likely to be achieved. Recaptured animals will be dispatched humanely.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	The purpose is to study the behaviour of wild grey squirrels in relation to efforts to detect and trap them.
2. Reduction Explain how you will assure the use of minimum numbers of animals	The number tagged will be constrained by practical considerations to a maximum of 50 per annum. This is likely to give a relatively imprecise estimate of detectability, but of greater interest is the insight given into how individual squirrels respond to the choice of feeding points offered by the detector array.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	The tagging method used is designed to be benign, and has already been used in many squirrel studies without adverse effects being reported. However, in tagging any wild animal some risk of adverse effects must be acknowledged. In our experimental design, animals will be tagged for only a few weeks before recapture and dispatch, minimising any risk of poor welfare occurring. Poor condition of animals on recapture, or the unexplained disappearance of tagged individuals during the study, will be taken as suggestive of a problem with the tagging method.

PROJECT 2	**Positive exclusion screen trials for fish**	
Key Words (max. 5 words)	Salmon, Eel, Vertical bar screen	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3)	X	Basic research
		Translational and applied research
		Regulatory use and routine production
	X	Protection of the natural environment in the interests of the health or welfare of humans or animals
	X	Preservation of species
		Higher education or training
		Forensic enquiries
		Maintenance of colonies of genetically altered animals
Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)	The objective of the study is to assess how effective 10 and 12.5mm vertical bar screens are in deflecting downstream migrating Atlantic salmon (*Salmo salor)* smolt and European eel (Anguilla anguillo*).* A screen positioned within a flume will guide fish moving downstream to a bywash channel located at the downstream end of the screen. Underwater camera arrays located immediately downstream of the bywash and behind the screen will enable an accurate count of fish passing via each route. The attachment of an acoustic transmitter to smolt and eel will allow the fine scale movement of tagged fish in each release group to be observed in two dimensions (2D) and mapped within a GIS system.	
What are the potential benefits likely to derive from this project (how science could be advanced or	The potential benefit of this study will be improved regulatory standards for screen design at water undertakings to ensure the protection of migratory fish and	

humans or animals could benefit from the project)?	improvements in screen and bywash design to prevent injury and loss of migratory fish whilst maximising the hydraulic performance of the screen.
What species and approximate numbers of animals do you expect to use over what period of time?	We aim to attach acoustic transmitters to a total of approximately 440 Atlantic salmon smolts and 440 European ccl (silver) over the duration of the study.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	The external attachment of a tag weighing no snore than 2% of the mass of the fish for a relatively short period of its natural downstream migration is believed to have a minimal negative impact on the fish in the study. The attachment procedure is considered to be of a minor severity. The fish will be maintained under general anaesthesia throughout the tag attachment procedure. If the use of external tags is considered to interfere with the normal transit of a fish through or along a screen which could result in either impingement of the fish upon the screen or loss of the tag then the tag will be attached internally requiting a surgical procedure. All surgical procedures have the potential to result in health problems for fish such as disturbance of physiological function, or more subtle behavioural or immunological effects. However following a short period of perturbation following capture and tagging no significant effects on swimming behaviour are typically observed in fish following this procedure. Handling stress will be improved by rendering the fish unconscious (anaesthesia) during the operating procedure. The procedure to attach the transmitter to the fish is considered to be at a moderate level of severity. Wild fish fitted with and external tag will be released back to the wild after the trial and following removal of the tag.

	All fish fitted with and internal tag and all captive bred fish will be recaptured after the trial by placing a f'ke net at the outlet of the flume and humanely killed in accordance with Schedule 1.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	The use of live animals is needed to confirm the currently unknown behavioural response of fish to various physical variables of a positive exclusion screen. Dead fish or inert objects would not show active behavioural component of the screen deflection process simply following the flow lines in front of the screen.
2. Reduction Explain how you will assure the use of minimum numbers of animals	The design of the study within an open channel will allow for the behaviour of naturally occurring wild fish (untagged) passing through the flume (trial arena) to be observed and quantified to provide a measure of the effectiveness of experimentally tagged fish to act as a surrogate, thus reducing the number of individuals required to be tagged.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	The fish species chosen arc species of conservation concern and the study will inform management and regulatory objectives designed to derive long-term conservation gains for these species. Wild and Captive bred fish will be tagged with external tags that are both quicker and less invasive to fit, potentially reducing the initial stress levels to the fish. The behaviour of fish within each trial will be observed to determine whether this method of tag attachment is fit for purpose or whether a method of internal attachment is required.

<table>
<tr><td>PROJECT 3</td><td>Improving models of species interactions by including individual heterogeneity</td></tr>
<tr><td>Key Words (max. 5 words)</td><td>Rodents, mustelids, population dynamics</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td>5 years</td></tr>
<tr><td>Purpose of the project (as in section 5C(3)</td><td>
<table>
<tr><td>Basic research</td><td>Yes</td><td></td></tr>
<tr><td>Translational and applied research</td><td>Yes</td><td></td></tr>
<tr><td>Regulatory use and routine production</td><td></td><td>No</td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td>Yes</td><td></td></tr>
<tr><td>Preservation of species</td><td></td><td>No</td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td></td><td>No</td></tr>
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</td></tr>
<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td>The object of the project is to investigate the population dynamics of free-living wild populations, in particular predators and their prey. It is unknown how the differences between individuals affect the population dynamics of predators and their prey.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?</td><td>The data collected will allow us to construct models that will allow the more accurate prediction of the dynamics of interacting species. Better models should allow better conservation or species management strategies to be developed.</td></tr>
<tr><td>What species and approximate numbers of animals do you expect to use over what period of time?</td><td>We propose to use the following species. As we are dealing with free-living wild populations, it is hard to give exact numbers.

Rodents(mice ~1,000, voles ~1,000)</td></tr>
</table>

	Mustelids(weasels ~100, stoats ~100)
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	Hypothermia has been reported as potential side effect of live trapping rodents, although this is rare. For mustelids, we are aware of no reported cases, although it is a possibility. Handling of the individuals may cause stress; although this is short-term and we will make every effort to reduce this. Blood/tissue samples will be collected from individuals on first capture. The level of severity is expected to be mild. In rare cases some individuals may experience haemorrhaging. Animals are released at the point of capture. The return of the wild animals to the wild poses no threat to public health or the environment.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	As we are investigating the population dynamics of wild populations, it is not possible to replicate the conditions in the laboratory. There is no existing data available that contains the information that we require.
2. Reduction Explain how you will assure the use of minimum numbers of animals	As we are looking at populations we need to maximise the number of individuals captured. We have found that the optimum trapping frequency is once a fortnight as this provides the data we need to collect with the least amount of effect of the populations.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	Both rodents and mustelids have fast generation times and exhibiting dramatic fluctuations in population size both geographically and through time. Rodents are a very abundant species that are easy to trap and mark. As rodents make up a sizable proportion of mustelids' diet, they make a suitable predator species to study. Ample food and bedding will be provided in the live traps. Processing will be kept to a minimum and the animals are released at the point of capture.

<table>
<tr><td>PROJECT 4</td><td>Investigating virulence of, amphibian host range of and the effectiveness of the amphibian host response to ranaviruses</td></tr>
<tr><td>Key Words (max. 5 words)</td><td>Infectious disease, conservation, amphibian, ranaviruses</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td>5</td></tr>
<tr><td>Purpose of the project (as in Article 5)</td><td>
<table>
<tr><td>Basic research</td><td>Yes</td><td></td></tr>
<tr><td>Translational and applied research</td><td>Yes</td><td></td></tr>
<tr><td>Regulatory use and routine production</td><td></td><td>No</td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td>Yes</td><td></td></tr>
<tr><td>Preservation of species</td><td></td><td>No</td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td></td><td>No</td></tr>
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</td></tr>
<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td>We seek to determine the host range and host-specific virulence of a panel of ranaviruses with the purpose of assessing risk to European host species. We also will investigate how a UK host species responds to ranavirus in the UK and if genetic adaptations to ranaviruses are effective.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?</td><td>Ranaviruses are increasing geographic range in Europe and are causing serious disease and population declines in several European amphibian host species. Several variants are circulating in Europe. Our work will allow us to identify which ranavirus types are risks to the various host species we will use. Further, our work will show us if UK common frogs are adapting to this emerging threat and if the adaptations are suitable to reduce the risk of host decline and extinction.</td></tr>
<tr><td>What species and</td><td>We will use 5 host species (common frog and toad,</td></tr>
</table>

approximate numbers of animals do you expect to use over what period of time?	alpine newt, and both common and Mallorcan midwife toads) and will use no more than 1000 animals per species and per life history stage over the 5 year period.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	Some animals will be toe clipped in the wild for genetic work and we expect no adverse effects from this procedure. These animals will be released back to the wild after clipping. Animals used for experimental work may experience substantial effects of disease. All experimental animals will be euthanized either once a humane endpoint has been reached or when the end of an experiment is reached.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	We cannot examine the impact of an infectious agent on a host without the use of animal experiments. Cell and tissue culture cannot capture the range of responses or allow us to ascertain virulence.
2. Reduction Explain how you will assure the use of minimum numbers of animals	We have extensive experience with amphibian infectious disease experiments and have used our previous experience to calculate a maximum sample size per treatment of 40 $\pm$ 5 when host responses are subtle, 30 $\pm$ 5 when they are not.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	We have selected species at risk to ranaviruses in Europe, including UK natives and one non-native invasive species. We have extensive experience with stocking densities of larvae, juveniles and adults of all species listed, as well as feeding requirements and habitat enrichment. We minimize suffering in all species through strict adherence to euthanasia requirements, based on clearly identified humane endpoints.

<table>
<tr><td>PROJECT 5</td><td colspan="3">Chemicals and fish Regulation</td></tr>
<tr><td>Key Words (max. 5 words)</td><td colspan="3">Toxicity testing, environmental pollutants</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td colspan="3">Five years</td></tr>
<tr><td>Purpose of the project (as in Article 5)</td><td colspan="3"><table>
<tr><td>Basic research</td><td></td><td>No</td></tr>
<tr><td>Translational and applied research</td><td></td><td>No</td></tr>
<tr><td>Regulatory use and routine production</td><td><u>Yes</u></td><td></td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td><u>Yes</u></td><td></td></tr>
<tr><td>Preservation of species</td><td></td><td>No</td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td></td><td>No</td></tr>
</table></td></tr>
<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td colspan="3">To determine the acute and/or chronic toxicity of chemicals or formulations of chemicals intended for use in the aquatic environment or that may reach the aquatic environment as a result of application within a process, accidental spillage or malicious action.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?</td><td colspan="3">The key benefit is for protection of the environment from the detrimental effects of an increase in the concentration of chemicals. These chemicals may have the potential to impact on aquatic species and human health as a consequence of contact or ingestion of the water or of aquatic organisms that have been in contact with the water</td></tr>
<tr><td>What species and approximate numbers of animals do you expect to use over what period of time?</td><td colspan="3">Over the five year study period it is possible that a number of different fish species may be tested either as standard laboratory species or according to their relevance to a particular pollution issue that is being investigated. Up to fifteen hundred fish could be tested over this period</td></tr>
</table>

In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	Fish will be exposed to a single concentration or concentration range of a chemical to determine toxicity and to inform the setting of regulatory guidelines or appropriate use or application concentrations to avoid environmental impact. Some fish may die but the majority will experience lesser effects. Infrequently some fish will be anaesthetised and tagged to aid identification during a study. Adverse effects from this procedure are expected to be minimal as the procedure is conducted under anaesthesia. All fish will be humanely killed as soon as possible to avoid unnecessary suffering including those remaining at the end of a study. Procedures will be conducted so as to minimise the number of fish used and observations will be conducted sufficiently frequently that fish can be killed humanely as soon as there is evidence of adverse behaviour indicative of the onset of mortality..
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	Invertebrate or plant species toxicity data will be used to indicate an appropriate limit concentration at which to test a limited number of fish to confirm whether they are more sensitive. Only where data indicate greater sensitivity of fish will a more extended toxicity test with fish be used.
2. Reduction Explain how you will assure the use of minimum numbers of animals	All experimental work will utilise the published literature and previous experience by the Project Licence holder and colleagues who undertake similar work to ensure that the minimum number of animals are used that will permit a robust statistical and meaningful analyses of the results.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs	The purpose of the work is to provide advice on the conservation and management of fish stocks. Therefore, a range of fish species are likely to be used to reflect sensitivity of natural populations. The methods chosen are based on previous experience and research and will provide evidence that will form the basis of suitable advice primarily to industry on the factors affecting fish populations

(harms) to the animals.	and recommendations for suitable mitigation. Fish will be humanely killed at the first signs of adverse effects and any remaining fish upon study termination.

PROJECT 6	**Biological Effects of Hazards in the Environment**	
Key Words (max. 5 words)	Behaviour, cancer, neurodegeneration	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3)	X	Basic research
	X	Translational and applied research
		Regulatory use and routine production
		Protection of the natural environment in the interests of the health or welfare of humans or animals
		Preservation of species
		Higher education or training
		Forensic enquiries
	X	Maintenance of colonies of genetically altered animals
Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)	People are increasingly exposed to a broad mixture of electromagnetic fields in the environment, both at home and work. Some of these are generated by power lines and electrical appliances, and others by mobile telephones and base stations. While there are exposure guidelines to limit exposure both at home and at work, there are persistent concerns that adverse effects may occur at exposures well below these guideline values. These include increased risks of some types of cancer, and changes in brain function leading to effects on memory and even neurodegeneration and dementia. Our previous work is reassuring in that it does not suggest exposure causes health effects, but these new studies will address these possibilities further, and so help to provide more definitive answers.	
What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the	The information gained in this project will help to characterise more fully the potential of these fields to cause biological effects and will contribute towards an improved assessment of the health risks associated with exposure. The results will be published in peer-reviewed scientific journals and will contribute to filling gaps in knowledge about the health consequences of exposure.	

project)?	
What species and approximate numbers of animals do you expect to use over what period of time?	Mice. The team of eight scientists and technicians will work on this project and we expect to use approximately 3000 animals in total over the 5-year period. This is based on our experience of similar experiments.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	With the exposures and testing procedures we will use, we do not expect to see any adverse effects, only very minor or subtle changes. Nevertheless, all experimental animals will be carefully observed for signs of stress or illness. The level of severity is moderate to encompass possible adverse effects, particularly those associated with ageing in mice. The animals will be culled at the end of the experiment.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	It is not feasible to use in vitro models, non-sentient alternatives or to use invertebrates in these studies. We need to use alert, behaving animals that show a complete repertoire of physiological functions. Overall, the data obtained with such models are more valid for health risk assessment than that provided by any alternative models and allow a clearer extrapolation to the potential risks in humans.
2. Reduction Explain how you will assure the use of minimum numbers of animals	The numbers of animals used will be reduced as far as possible following specific advice from statisticians to maximise the amount of information obtained during an experiment and to minimise the numbers of animals used. We follow a system of written operating procedures to ensure consistency and reproducibility in the results and help to reduce unnecessary variability. In turn these should help to reduce the numbers of animals needed. The use of healthy animals, obtained from a reputable registered supplier, should also help to avoid loss of experimental animals through disease. We will use specialised video recording to monitor and record the responses of the animals and this will help to significantly increase the potential data return for each animal used.

3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	Mice have been chosen for these studies because they provide a very good model system to study neurobiology and carcinogenesis. There is a wealth of biological data concerning mice, and we have extensive experience of using this species in similar studies for many years. Furthermore, using mice maximises the potential for use of genetic markers in molecular studies and the potential for interpretation of results using genetic databases. None of the procedures proposed in these studies is anticipated to cause any animal suffering or harm but if any deviation from normal is noted following any treatment or assessment, the animal will be monitored closely, and receive additional care as necessary.

<table>
<tr><td>PROJECT 7</td><td>Diadromous & freshwater fish: assessment & ecology</td></tr>
<tr><td>Key Words (max. 5 words)</td><td>Fish, Stock Assessment, Ecology, Environment, Conservation.</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td>5</td></tr>
<tr><td>Purpose of the project (as in section 5C(3)</td><td>
<table>
<tr><td>Basic research</td><td></td><td>No</td></tr>
<tr><td>Translational and applied research</td><td></td><td>No</td></tr>
<tr><td>Regulatory use and routine production</td><td></td><td>No</td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td>Yes</td><td></td></tr>
<tr><td>Preservation of species</td><td>Yes</td><td></td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td></td><td>No</td></tr>
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</td></tr>
<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td>1. Estimate natural marine mortality rates, any trends in natural mortality, and the level of fishery exploitation for specific salmon and sea trout stocks.

2. Investigate how the environmental conditions experienced by juvenile and adult salmonids, as well as other diadromous species (e.g. eels and lamprey), may impact them at both the individual and population levels.

3. Determine the risks and impacts of introduced fish/ crustacean species (non-native, translocated, re-introduced) on native fish populations, including factors associated with a changing climate.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or</td><td>Information on all aspects of fish mortality/ exploitation rates, abundance, distribution, migration, ecology and the impact of introduced fish species under a changing environment will ensure</td></tr>
</table>

animals could benefit from the project)?	that the best available information is available to support Policy in implementing fish management and conservation measures. The information will also contribute to the development of methods for assessing Atlantic salmon and European eel stocks.
What species and approximate numbers of animals do you expect to use over what period of time?	Diadromous, freshwater and introduced fish species. Approximately 93,000 fish over the 5 year duration of the Licence.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	The vast majority of fish (approximately 99%) will be released back to the wild, with no predicted adverse effects, following procedures with a mild level of severity.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	The principal aim of the work is to determine mortality rates or describe the behaviour and ecology of fish in relation to changes in their natural aquatic environment in order to conserve and manage populations. Therefore, there is no alternative to the use of living animals.
2. Reduction Explain how you will assure the use of minimum numbers of animals	All experimental work will utilise the published literature and previous experience by the Project Licence holder, and his/ her colleagues who undertake similar work, to ensure that the minimum number of animals are used that will permit a robust statistical and meaningful analyses of the results. All experimental work will be discussed and agreed with a professional statistician who advises on statistical analyses. The statistician will provide statistical support to all aspects of the research, from designing the experimental approach to conducting and reporting the analyses.
3. Refinement Explain the choice of species	The purpose of the work is to provide advice on the conservation and management of fish stocks. Therefore, a range of fish species (including

and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	salmon, sea trout, European eels, coarse fish, and non-natives) need to be studied in order to provide relevant and meaningful data to support Policy and management decisions. The research will produce information and data on mortality & exploitation rates and the impacts on fish populations from a wide range of environmental and man-made changes to the aquatic environment. The methods chosen are based on previous peer-reviewed research and experience and will provide evidence that will form the basis of suitable advice to Government on the factors affecting fish populations and recommendations for suitable mitigation. Where fish undergo a procedure and recovery, they will be monitored for a suitable period of time in order to assess any adverse impacts and ensure a minimum of suffering.

<table>
<tr><td>PROJECT 8</td><td>Ecology of freshwater fish</td></tr>
<tr><td>Key Words (max. 5 words)</td><td>Fish, non-native fish, habitat loss, eutrophication</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td>5</td></tr>
<tr><td>Purpose of the project (as in Article 5)</td><td>
<table>
<tr><td>Basic research</td><td>Yes</td><td></td></tr>
<tr><td>Translational and applied research</td><td>Yes</td><td></td></tr>
<tr><td>Regulatory use and routine production</td><td></td><td>No</td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td>Yes</td><td></td></tr>
<tr><td>Preservation of species</td><td></td><td>No</td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td></td><td>No</td></tr>
</table>
</td></tr>
<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td>The objectives are to:

1. Determine the impact of non-native fish on freshwater ecosystems and freshwater fish communities
2. Determine the consequences of nutrient enrichment, habitat loss and climate warming on freshwater fish communities
3. Identify the cumulative impacts of the multiple stressors of climate warming, habitat loss, nutrient enrichment and non-native fish on freshwater ecosystems and freshwater fish communities
Each objective addresses a significant scientific unknown.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the</td><td>The potential benefits are the derivation of scientific knowledge that is used to develop policy and procedures that better regulate the management of rivers in the UK for the benefit of freshwater ecology and biodiversity.</td></tr>
</table>

project)?	
What species and approximate numbers of animals do you expect to use over what period of time?	The species are fish encountered throughout UK freshwaters, including some non-native species. These are Roach *Rutilus rutilus*, common bream *Abramis brama*, rudd *Scardinius erythrophthalmus*, dace *Leuciscus leuciscus*, tench *Tinca tinca*, brown trout *Salmo trutta*, Atlantic salmon *Salmo salar,* European eel *Anguilla anguilla*, crucian carp *Carassius carassius*, bullhead *Cottus gobio*, gudgeon *Gobio gobio*, 3 spined stickleback *Gasterosteus aculeatus*; minnow *Phoxinus phoxinus;* Common carp *Cyprinus carpio,* gold fish *Carassius auratus;* sunbleak *Leucaspius delineatus*, topmouth gudgeon *Pseudorasbora parva*, fathead minnow *Pimephales promelas*, pumpkinseed *Lepomis gibbosus*; black bullhead *Ameiurus melas*; European catfish *Silurus glanis*; zander *Sander lucioperca*; grass carp *Ctenopharyngodon idella;* European barbel *Barbus barbus*; chub *Leuciscus cephalus* Over the 5 year project, a maximum of 5000 fish will be used. An individual fish is likely to spend no more than 100 days under a Protocol.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	The expected adverse effects are inhibited growth rates, shifts in behaviour and modifications to the diet of the fish. In conjunction with these are the use of anaesthesia, insertion of tags for identification and the effects of environmental manipulations. The expected level of severity is mild in all protocols, other than protocol 6 (moderate). The welfare of the fish is measured according to a scoring of the behaviour of the fish during daily observations. This is in relation to their respiration, feeding, swimming behaviour and response to external stimuli. Severity is also measured according to the weight loss of an individual fish (maximum of 10 % loss for all protocols) The animals will either be killed at the end or released back into the wild where they have been

	used at a POLE.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	Non-animal alternatives cannot be used in the project as the work is specific to investigating how different stressors impact the ecology of UK fish species. This requires measurement of their behaviour, diet and feeding interactions and growth rates, all of which require the use of live fish.
2. Reduction Explain how you will assure the use of minimum numbers of animals	Minimum numbers of animals will be used in the project by ensuring each experiment if designed with the advice of a statistical expert. That expert will advise on the number of treatments, replicates and fish being used, with multi-factorial designs used where possible to minimize numbers. The aim of the statistics is to indicate the minimum number of animals required to provide statistically robust data.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	The fish used in the project are all present in UK freshwaters, with their status as either native or non-native, and have been identified as being important ecologically by regulatory authorities. Consequently, they are refined for use in the project in order to meet the project objectives. To minimise harm, the fish to be used experimentally will be sourced from aquaculture facilities, with the Environment Agency's fish farm facility used wherever possible where the fish are maintained to high husbandry standards that are usually free from any confounding factors, such as parasite infections. Animal handling will be minimised to periods of data collection when they will be under general anaesthetic. Where temperature and environmental conditions are being varied within a trial then these will be within the range of those experienced by the animals in natural situations. Where fish are being tagged using Passive Integrated Transponder tags or elastomer tags, and/ or a tissue biopsy is being taken, then these

	procedures will be completed on the fish under general anaesthetic using MS-222. Appropriate periods of acclimatisation to tank and pond conditions will be used and where tagging is used to monitor fish performance, those used will be the most appropriate and least invasive. The use of a behavioural and weight loss scoring system within daily observations of fish in tank and pond conditions also minimises welfare costs to the animals through clear definition of endpoints. In work completed at POLES, fish will only be released back into the wild once they have recovered from procedures, as demonstrated by their response to external stimuli, ability to swim and to maintain their normal body position.

PROJECT 9	**Evaluation of rodenticides in the laboratory and field.**	
Key Words (max. 5 words)	Rodenticides, rodents, resistance, anticoagulants.	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3)		Basic research
	X	Translational and applied research
	X	Regulatory use and routine production
	X	Protection of the natural environment in the interests of the health or welfare of humans or animals
		Preservation of species
		Higher education or training
		Forensic enquiries
		Maintenance of colonies of genetically altered animals
Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)	This Project Licence will authorise work to monitor, regulate and develop rodent control methods, to monitor the environmental impact of chemicals used to control rodents, and to investigate the development of physiological resistance to the anticoagulant rodenticides, which are the rodent control compounds most widely in use today.	
What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?	Rodents can be a major pest to man, because they consume our food, they are involved in the transmission of many diseases, they damage our property through their continual requirement to gnaw, and they can be a major conservation problem, particularly on oceanic islands where they have been responsible for the extinction of many endemic species of birds. The development of the anticoagulant rodenticides in the early 1950's revolutionized rodent control, providing highly effective poison baits with a delay between consumption of a lethal dose and the onset of symptoms. This delay prevented the rodents associating cause and effect, and	

	thus prevented the development of “bait shyness” or “conditioned bait aversion”; the main drawback of most other “acute” rodenticides. However, the recent development of practical resistance to the more toxic anticoagulants that are registered for use in and around farm building is now a major problem in the UK, Germany, France and elsewhere. The research authorised by this Project Licence will be used to: • extend the useful life of anticoagulant rodenticides • investigate resistance to anticoagulant rodenticides and develop resistance management strategies • investigate the non-target impact of the anticoagulant rodenticides, particularly on bird species, and to develop methodologies and formulations that will minimise this impact • develop more effective rodenticide formulations • develop novel rodenticide active ingredients The output from this research is currently being used by the UK Regulatory Authorities (Health and Safety Executive) and the European Commission, and has benefited developing countries, where the rodent problem has life threatening impacts on subsistence crop production, and where the potential impact against species of conservation concern is far greater.
What species and approximate numbers of animals do you expect to use over what period of time?	The output from this research is currently being used by the UK Regulatory Authorities (Health and Safety Executive) and the European Commission, and has benefited developing countries, where the rodent problem has life threatening impacts on subsistence crop production, and where the potential impact against species of conservation concern is far greater. For the regulation and development of rodenticides approximately 4000 Norway rats and 3800 House mice will be used over the five year period. To investigate the development of physiological resistance to the anticoagulant rodenticides, approximately 2000 Norway rat and 2000 House mice will be used over the five year period. To investigate the environmental impact of anticoagulant rodenticides, approximately 150 pigeon, 150 quail, 150 Norway rats and 150 House mice will be used over the five year period.

	For the investigation of physiological resistance it is necessary to conduct studies on rats and mice trapped from wild populations.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	In recent severe test Protocols that involved anticoagulants: • 14.2% of Norway rats and 12.5% of house mice were humanely killed with no record of the animals developing adverse symptoms. • 43.6% of Norway rats and 53% of house mice were humanely killed because they had developed symptoms that were an end-point to the Protocol • 42.1% of Norway rats and 34.5% of house mice were found dead as a result of the Protocol. In future mortality tests with anticoagulants, an important objective will be to reduce the proportion of Norway rats and house mice that are found dead as a result of the Procedure, and increase the proportion of animals that are humanely killed because they have reached the end point to the Procedures. Currently, the majority of work is conducted on anticoagulant rodenticides, and the most common symptoms are anaemia in both species, laboured breathing (mainly in mice) and conjunctival anaemia (mainly in rats). Less common symptoms in rats (and very occasionally in mice) are partial loss of hind limb control and obvious swellings around the head, shoulders or legs. External bleeding can occur in rats (more frequently in resistant strains than albino strains) and less frequently in mice. For resistance studies, the animals will be dosed with anticoagulant, and between 90% and 95% of them will be killed before the animals develop any adverse symptoms. For the environmental impact work, the Norway rats and House mice will be dosed with anticoagulant whilst at the same time being given vitamin K1 (the complete antidote to the anticoagulant), until they are no longer affected by the anticoagulant (i.e. do not have a prolonged clotting time 24 hours after the vitamin K1 has been removed). Subsequently, the animals will be humanely killed, and their livers removed for subsequent anticoagulant residue analysis. The pigeons and quail will be dosed with anticoagulant by intraperitoneal injection, and will be killed before the animals develop any adverse symptoms. Blood samples taken by cardiac puncture under terminal anaesthesia, to determine their sensitivity to the anticoagulant.

Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	For regulatory studies, there is no alternative to using live animals, as detailed Government guidelines (from the UK, Europe, the US and elsewhere) specify the animal species and group size required for data submission. For the investigation of physiological resistance there is no alternative to using live animals. The research usually involves the use of wild caught Norway rats and House mice from locations where the animals are causing a significant problem, often where commercial control procedures have failed. To investigate the non-target impact of anticoagulant rodenticides on mammals and birds, there is no alternative to the use of live animals. Where possible, the procedures of a 'moderate' severity limit will be conducted on pigeon and quail. In the past, pigeons have been obtained from people that race pigeons, utilizing their surplus of birds that would otherwise be killed. Both pigeon and quail could also be obtained from an approved animal suppler although the use of pigeons that would otherwise be killed would be preferable (replacement).
2. Reduction Explain how you will assure the use of minimum numbers of animals	Efficient experimental designs will be used to minimise the required sample size whilst maintaining adequate precision. Where appropriate, advice from a professional statistician will be sought to ensure the number of animals required is minimised. Further, when necessary, pilot studies will be used in the design of a main study, to ensure the best information possible is used in its statistical design, ultimately reducing the number of animals required. For the anticoagulant rodenticides a number of end points to the Protocol have been identified that are now implemented routinely in order to reduce unnecessary suffering. The acceptance of these end points by the UK regulatory authorities (The Health and Safety Executive) is an important objective of this Project Licence.
3. Refinement Explain the choice of species and why the animal model(s) you	The Norway rat and House mouse are two globally important rodent pest species, and are the main target species for rodenticides and rodent control. The requirement to conduct tests with a severe severity limit, and the use of the proposed animal models are specified by the Regulatory Authorities in Europe, the USA, Australia

will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	and elsewhere. When developing products, Sponsors require the work to be conducted using the animal models and protocols specified by the appropriate Regulatory Authorities. The anticoagulant rodenticides achieve their effect by compromising the coagulation system, thus causing a lethal haemorrhage which can occur between 3 and 15 days after consumption of a lethal dose. The location of the haemorrhage is variable and the resulting mortality can occur suddenly, without the development of obvious symptoms, or the animal can develop symptoms over several days, and will be humanely killed after developing the humane end points to the protocol. For resistance testing, a new molecular technique has been developed that can be used to assess physiological resistance in Norway rats and house mice, without the requirement to conduct tests on live animals. Proposed work will be conducted to validate the molecular methodology, by measuring the magnitude of the resistance for each of the resistance mutations identified to date, in order to assess their likely impact of the resistance on treatment outcome. To do this it is necessary to use live animals. To this end, new resistance testing methodologies have been developed at this laboratory that are authorised by Project Licence Protocols with a 'mild' severity limit. All previously published resistance tests for the second generation anticoagulants are authorised by Project Licence Protocols with a 'severe' severity limit. Today the vast majority of resistance testing is conducted without the use of live animals. The applicant is currently in discussions with organisations in the US, Brazil, Mexico, Russia, Hungary, Czechoslovakia, and the Philippines, with the objective of extending the new molecular methodology into other territories. For all animals testing that will be regulated by this Project Licence application, experienced technicians will continue to record symptoms that develop in the test animals, so that end points to the Procedures can be refined to minimise suffering.

PROJECT 10	**West Country salmon and sea trout migration project**	
Key Words	Salmon, sea trout, smolts	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3)		Basic research
		Translational and applied research
		Regulatory use and routine production
	X	Protection of the natural environment in the interests of the health or welfare of humans or animals
		Preservation of species
		Higher education or training
		Forensic enquiries
		Maintenance of colonies of genetically altered animals
Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)	Due to the importance of the Region's rivers for salmonids, and the flow issues and barriers to migration and impoundments (any structure that holds the water back from its natural flow regime), there is a necessity to examine rivers in relation to smolt and adult numbers, survival and migration in order to understand the specific population dynamics and associated bottlenecks, to move European Union Framework Directive (WFD) classification of such rivers toward *'Good Ecological status'* and provide a sustainable healthy fish stock. Due to uncertainties at sea, and parasitic occurrences and the concomitant negative effects upon health and survival, the status of salmonids in freshwater are even more important to understand and improve survivability of the species. Therefore, the study proposes to examine both ends of the life stages of salmon and sea trout, the migrating juveniles and the returning adults	

	Objectives are to: • Assess impact of abstraction (taking water for irrigation and drinking water supplies) and flow conditions that influence adult and smolt migration, • Determine targeting of flow releases where possible and practicable • Determine extent of migration of adult fish.
What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?	Determination of the extent of barriers, impoundments and abstraction points (such as weirs, structures that contain pumps for the abstraction of water on a river flow regimes) that affect salmon and sea trout smolt migration, can lead to resultant amelioration of these effects though management actions to ensure that an overall improvement of smolt migration is achieved e.g. through change flow regimes or remedial works to barriers or impoundments. This is considered to provide a significant contribution to enhancing depleted salmonid stocks, which is important for the local and national economy. The project aims to identify the impact that barriers and impediments to migration can have on migration and returning adults. Information gained will be used to inform future river management practices, aiding greater success in the levels of returning adults and hence positively influence population levels. Knowledge gained from the identification of the potential benefits achieved from compensatory discharges can be utilised in other rivers management plans as appropriate.
What species and approximate numbers of animals do you expect to use over what period of time?	350 Salmon and sea trout/ year. In a Five year study this will be 1750 fish
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	Wild fish will be captured and anaesthetised in order to carry out surgery to place tags into the fish, prior to them being re released. This will allow the monitoring fish movements remotely from a series of tracking stations eg. radio antennae located along the river. Expected adverse effects are very low *(<0.5%).* However, there is the potential for some fish to be damaged during capture and surgery, (a rare event) and if so, and deemed not likely to recover, they will

	be humanely killed.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	Assessment of flow rates, compensatory flows, barriers and abstraction points could potentially be carried out superficially by comparing peer reviewed papers. However, since each river, flows and barriers or abstraction points presents its own unique set of problems, any such comparison would only be an *in principle,* relating to a type of barrier or abstraction point, but would still require assessment in order to determine extent of migration or escapement. Furthermore, - assessment of entrainment in abstraction sites can only be evaluated on a site specific basis. Determination of migration or escapement rates can only be achieved by monitoring salmon and sea trout on their specific river. Requires site specific assessment.
2. Reduction Explain how you will assure the use of minimum numbers of animals	Numbers of salmon and sea trout smolt required for tagging have been set at 350 per year, per river. The estimate accounts for greater numbers of smolts than adults, which is reflective of most Westcountry rivers population dynamics. The estimate of numbers of tagged fish is generated from data from neighbouring comparable rivers provided by statutory bodies and NGO's because there is no population data available for the e at present. However, it must be borne in mind that the numbers tagged are likely to be reflective of the population density, and therefore a proportion of the estimated population may be used if numbers are low, but total numbers would not exceed 350 fish per year per river.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	The project is required under EU directives (WFD) and Better Sea Trout and Salmon Fisheries 2008 - 2021 (EA). Catching and tagging salmon and sea trout will enable tracking of smolt migration past barriers and abstraction points, and assessment of their passability as well as assessing the effectiveness of works designed to help sea trout and salmon. Capture of adults returning to spawn will enable tracking of their migration in freshwater and identification of barriers to this migration. Tagging fish to monitor their movement remotely, using equipment such as radio antennae or sonar

	receivers is a well-established methods used internationally, and has a proven record of providing this kind of information, with minimal risk to the fish. The applicant has extensive experience of sonar (hydroacoustics) and radio frequency identification tagging a variety of fish species including salmon and sea trout, minimising risks and potential mortalities through good technique and relevant experience. Harm to the fish is minimised by aseptic surgical technique when placing tags. Anaesthesia is kept shorts and fish are monitored to ensure they are fit for release.

PROJECT 11	**The population ecology of North Sea seabirds**	
Key Words	Seabirds, climate change, diet, physiology, demogaraphy	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3)		Basic research
		Translational and applied research
		Regulatory use and routine production
	X	Protection of the natural environment in the interests of the health or welfare of humans or animals
		Preservation of species
		Higher education or training
		Forensic enquiries
		Maintenance of colonies of genetically altered animals
Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)	One of the most important measures of the conservation status of a species is its population size, and the dynamics of populations has long intrigued biologists. Population size is the product of birth rates, death rates, immigration and emigration (collectively known as demographic rates). A range of interacting processes all play an important role in determining these rates, including food supply, competing species, enemies (predators and parasites) and physical processes such as weather conditions. Factors associated from human activities are critically important in determining demographic rates. In particular, climate change has become a huge concern in recent decades because of dramatic effects on ecosystem dynamics and species health. Other man-induced impacts may also be important, such as invasive species, pollution and human developments. Intrinsic mechanisms are also believed to be important in regulating population size. Among colonial species, competition for food drives the relationship between population size and feeding rates to offspring. Genetic diversity is also regarded as an integral part of the sustainability of wild populations. Those species that have gone through population bottlenecks have a greatly reduced genetic diversity, which is predicted to make them more vulnerable to extinction. Environmental and intrinsic	

	drivers of change in population size are mediated via changes in the behaviour and physiology of individuals. Thus, studying individual behaviour and physiology is critically important to quantifying the mechanisms underlying changes in demographic rates which, in turn, determine population size. Commencing in 1973, the CEH's long-term study of seabird populations on the Isle of May (IMLOTS) and associated colonies has grown to be one of the most data-rich and complex studies of its kind in the world. Many species of seabird have declined over the last two decades, as a result of climate change, fisheries and other factors. Currently, there is also a concern that the expansion of marine renewable developments may also have a detrimental effect on seabirds. Seabirds are legally protected, so it is necessary to understand these drivers of change in their populations so we can support UK coastal economies whilst minimising environmental impacts. Therefore, the overall aim of the project is to quantify the impact of climate and other human-induced change on seabird populations. To achieve this aim, there are three elements of research that that fall within the jurisdiction of the Animal (Scientific Procedures) Act 1986: a) to quantify diet and foraging performance of seabirds; b) to estimate the effects of pollutants and parasites on seabirds; c) to quantify gene flow. By combining these methods with other research methods we are able to maximise our understanding of the causes of year-to-year variation in North Sea seabird populations, enabling us to provide high quality advice to policy makers, conservation managers and industry.
What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?	Understanding the drivers of seabird population change is beneficial because the UK holds internationally important numbers of numerous species and has a legal obligation under the EU Birds Directive to protect them. To understand the interacting effects of drivers of seabird populations requires detailed research to build up a firm evidence base, which is required to design appropriate and effective marine planning and conservation management strategies. CEH's long-term study of seabird populations on the Isle of May has provided much of evidence on the effects of climate change, fisheries and marine renewables on North Sea seabirds that policy makers and industry require to deliver sustainable use of UK coastal waters by integrating economic and conservation requirements.

What species and approximate numbers of animals do you expect to use over what period of time?	European shag (approx 2000/year) Black-legged kittiwake (approx 400/year) Common guillemot (approx 400/year) Razorbill (approx 400/year) Atlantic puffin (approx 400/year) Northern fulmar (approx 400/year)
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	The likely severity is mild. We expect birds to experience some short term mild discomfort lasting no more than a few minutes, and birds will be returned to the wild to continue their normal activities. Many years of experience of carrying out research of this kind has shown that adverse effects do not occur and birds return to normal behaviour quickly.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	The effects that we wish to measure on the physiology and behaviour of individual seabirds can only be measured on individuals living in the wild. There is no method of capturing the same information in vitro.
2. Reduction Explain how you will assure the use of minimum numbers of animals	We use the minimum number of animals that provide us with the statistical power to test our scientific questions. Setting this minimum value is based on our long experience of undertaking research of this kind, and on power analyses that have been carried out on past data.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general	The rationale for working on these seabird species is that they are good model species for understanding the drivers of seabird populations. Furthermore, the populations on the Forth Islands, in particular on the Isle of May, have been studied since 1973 so we have a substantial knowledge of their ecology and a strong background in identifying the drivers of population changes in these species. Seabirds are an ideal system for establishing the link between environmental change and species' health by studying the

measures you will take to minimise welfare costs (harms) to the animals.	physiology and behaviour of individual animals because they show minimal effects of being caught and handled by people, and respond well to procedures such as those set out in this application. Furthermore, we continually seek to minimise animal suffering by reducing handling time to a minimum and adopting handling and procedural methods based on our long experience with the species in question.

<table>
<tr><td>PROJECT 12</td><td>Novel Rodenticide Project</td></tr>
<tr><td>Key Words (max. 5 words)</td><td>Rodents, Rodenticides, control, toxicity</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td>5</td></tr>
<tr><td>Purpose of the project (as in section 5C(3)</td><td>
<table>
<tr><td>Basic research</td><td>Yes</td><td>No</td></tr>
<tr><td>Translational and applied research</td><td>Yes</td><td>No</td></tr>
<tr><td>Regulatory use and routine production</td><td>Yes</td><td>No</td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td>Yes</td><td>No</td></tr>
<tr><td>Preservation of species</td><td>Yes</td><td>No</td></tr>
<tr><td>Higher education or training</td><td>Yes</td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td>Yes</td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td>Yes</td><td>No</td></tr>
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</td></tr>
<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td>The aim is to undertake research and development and generate efficacy data on new and novel actives for the control of pest species of rodents (rats, mice). Existing rodent control relies heavily on anticoagulant rodenticide baits. However, important concerns for the long-term viability of these techniques make the search for new and improved actives a matter of high priority. Also, resistance to anticoagulants in Norway rats and house mice is a significant problem in many countries and therefore, any novel rodenticides discovered would have significant global benefits.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?</td><td>Research into improved actives for rodent control will benefit human, animals and the environment. This is because there is a need to improve safety to non-target species and find actives with a safer environmental profile to reduce environmental contamination. This research necessitates animal testing, and is justifiable at three levels. Firstly, the basic requirement to control rodents because of</td></tr>
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	their pest status. Secondly, that existing control methods have a variety of difficulties and failings that make improved actives highly desirable and thirdly, to fulfil regulatory requirements.
What species and approximate numbers of animals do you expect to use over what period of time?	Rats and mice. In the laboratory approximately a total of between 100 and 400 rats plus mice a year depending on the success of the research.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	Overall death is expected in approximately 50% of animals used. However, it is expected that 60% of all animals recorded as dead will have been culled according to humane endpoints by a Schedule 1 method.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	Rodenticides are designed to be mammalian toxicants and are thus a unique group of biocides/pesticides. The ultimate test of efficacy is lethality and this must be assessed against the target organism. It follows, therefore, that animals cannot be replaced by experimental models.
2. Reduction Explain how you will assure the use of minimum numbers of animals	1. Using in-vitro methods when possible 2. using a logical progression through the studies so the maximum amount of knowledge has been obtained before the next study commences.. 3. using only 2 animals for initial investigations into a new active.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	1. Humane end-points are stringently employed. Any animal exhibiting clinical signs of a type and severity which may be expected to effect death is killed, by a Schedule 1 method. 60 per cent of all animals recorded as dead will be killed according to this criterion. 2. Also, throughout the work every effort is made to develop experimental methodology to use non-lethal end-points 3. Protocols allow starvation periods of up to 12 hours, for the vast majority of procedures, the starvation period is only 6 hours.

<table>
<tr><td>PROJECT 13</td><td colspan="3">IMPACTS AND MITIGATION OF RIVER STRUCTURES ON FISH</td></tr>
<tr><td>Key Words (max. 5 words)</td><td colspan="3">Dams, hydropower, hydrodynamics, acoustics, telemetry</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td colspan="3">5 years</td></tr>
<tr><td>Purpose of the project (as in Article 5)</td><td colspan="3"><table>
<tr><td>Basic research</td><td>Yes</td><td></td></tr>
<tr><td>Translational and applied research</td><td></td><td>No</td></tr>
<tr><td>Regulatory use and routine production</td><td></td><td>No</td></tr>
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<tr><td>Preservation of species</td><td>Yes</td><td></td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals</td><td></td><td>No</td></tr>
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<tr><td>Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)</td><td colspan="3">The primary objectives of this project are: 1) to quantify the impact of river structures (e.g. hydropower or marine turbine facilities, weirs, dams etc.) on fish; and 2) to develop mitigation technology to reduce the magnitude of the impact experienced.</td></tr>
<tr><td>What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?</td><td colspan="3">Information obtained will provide evidence of impact of river infrastructure on fish distribution, migration, and survival, and prioritise conservation actions based on understanding of areas of greatest impact. Information obtained will help regulators ascertain whether current mitigation options (e.g. fishways and screens) are effective for the species of interest, and highlight areas where limited resources should be directed to maximise improvements. Identification and quantification of secondary impacts associated with river</td></tr>
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	infrastructure and practices, such as the sounds created, will be achieved so that preventative measures can be developed, and so that alternative mitigation technologies may be advanced.
What species and approximate numbers of animals do you expect to use over what period of time?	Fish (including *Petromyzontidae, Anguillidae, Salmonidae, Thymallidae, Cyprinidae*). Total maximum number = 10,000 over 5 years
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	The expected mild to moderate adverse effects on fish will primarily relate to the capture, handling, and marking / telemetry procedures used. Mild adverse effects may also relate to use of anaesthetic and insertion of hypodermic needles e.g. to collect blood samples or implant visual markers.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	It is currently impossible to model migratory behaviour of fishes, or how non-migratory fish move within their environment, in response to environmental conditions encountered at riverine structure. Live animals must be used to obtain the information necessary to facilitate greater understanding of this so that models may eventually be developed. This project will provide information needed to develop a behavioural "rule base" based on empirical observation. The approach proposed will use the minimal amount of animals to obtain the information necessary to replace their use in the future by providing the information to help construct appropriate models.
2. Reduction Explain how you will assure the use of minimum numbers of animals	Calculations of statistical power of the research required to meet the stated objectives forms an iterative component of our project. Assessments of statistical power based on data collected in any one phase will form part of the decision making process in relation to selection of sample size in future phases. This way the number of fish required to ensure meaningful conclusions will be minimized.
3. Refinement Explain the choice of species	It is essential that the animals used are the same species for which this research is intended to benefit by developing the most appropriate

and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	management strategies based on real world observations. The species selected represent those that are of high conservation and/ or economic value as defined by national and international legislation, yet maintain populations that are relatively stable and healthy in the study rivers selected. The selected species also provide representatives of fish with a range of different body morphologies, swimming capabilities, behaviours, and life history traits. All surgical techniques will use well developed and widely used protocols to minimize handling and associated stress. Effects of the techniques will be monitored to reduce probability of causing pain and suffering during future phases. Behavioural traits will be monitored to indicate humane endpoints i.e. the earliest indicator in an animal experiment of severe pain, distress, suffering, or impeding death. Post-surgery behaviour will be closely monitored over the period of recovery to assess deviation from the pre-surgery condition. The data will be regularly reviewed to assess whether behavioural measures can be refined to enhance the efficiency of identification of humane endpoints based on a relationship between exhibition of aberrant behaviour and resulting deterioration in condition.