Animals (Scientific Procedures) Act 1986

Non-technical summaries for project licences granted during 2016

Volume 13

Projects with a primary purpose of maintenance of colonies of established genetically altered animals

Project Titles and keywords

1. **Breeding and Maintenance of Immunocompromised and Genetically Altered mice as a service**
    - Breeding, Immunodeficient, Genetically Altered

2. **The generation, maintenance and breeding of genetically altered mice as a service.**
    - Transgenic, Genetic Alteration, Breeding, Rodents

3. **Generation, Breeding and Maintenance of Genetically Altered Rodents**
    - Genetically altered, breeding, DNA, CRISPR

4. **Provision of a service for production and maintenance GA animals and antibody production**
    - Service, Genetically altered animals

5. **Producing and Maintaining Genetically Altered Rodents**
    - Breeding, Rederivation, Service, Transgenics, Rodents

6. **Breeding and Maintenance of Genetically Altered Rodents**
    - Breeding, Genetically Altered

7. **Creation, breeding and maintenance of genetically modified mice**
    - Mice, transgenics, knockout, cryo-preservation, rederivation

8. **Cryopreservation of Mouse and Rat Embryos**
    - Cryopreservation mouse and rat embryos

9. **Breeding genetically altered mice and rederivation**
    - Breeding genetically altered mice and rederivation

10. **Breeding and maintenance of GA mice**
    - Breeding, Maintenance, Genetically, Altered, Mice

11. **Breeding and Production of Genetically Altered Mice**
    - Genetic alteration, mouse, breeding

Project 1	**Breeding and Maintenance of Immunocompromised and Genetically Altered mice as a service**	
Key Words (max. 5 words)	Breeding, Immunodeficient, Genetically Altered	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3) (Mark all boxes that apply)	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)	The purpose of this project licence is to breed, maintain and supply high quality immuno-compromised and genetically altered mice for use in cancer research under the authority of other project licences at our establishment. In this way the breeding programmes can be managed in dedicated facilities and closely monitored to minimise overproduction and to produce a high quality healthy animal. Our experienced animal technologists supply the specific expertise necessary to provide the optimum husbandry conditions, the highest level of health and welfare as well as accurately maintaining the minimum colony size to produce the animals required for research. They consult closely with the Named Animal Care and Welfare Officer and the Named Veterinary Surgeon on any issues that arise in the colonies ensuring that any actions necessary are carried out promptly.	
What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could	As a direct result of the work that is carried out in our facility using these strains of mice, major and innovative developments in the treatment of breast, testicular, lung and gut cancers have been made in human patients. These have been widely publicised in	

benefit from the project)?	the media is recent years.
What species and approximate numbers of animals do you expect to use over what period of time?	Research into the causes and subsequent treatment of a wide range of cancer types requires the use of specific models of mice including the athymic nude, and animals with specific genetic modifications that are relevant to the genetic pathways now known to be associated with tumour development. We estimate that we will breed and use 4000 athymic nude mice each year and approximately 500 Genetically Altered mice per year for this 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?	The procedures on this project licence are all classified as Mild severity. We do not anticipate any adverse effects from the normal breeding of these animals. Any unexpected ill health or phenotypes will be managed promptly and the animals 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	Animals are used only where development and therapies are not possible in the laboratory. This licence aims to breed and maintain such animals and the detailed justification for the particular programmes of research is provided by the individual scientists in their own Licences that must always be approved by the Home Office.
2. Reduction Explain how you will assure the use of minimum numbers of animals	Careful management of breeding colonies will allow us to maintain the lowest number of breeding animals to produce the required number of study animals. Unfortunately due to the genetics of breeding Athyrnic Nude mice and some Genetically Altered mice, there will be surplus animals produced. These we will humanely kill at the earliest opportunity or use for collection of tissue and blood products for further use in research.
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.	Routine health screening of the existing colony and careful examination of the health status of any new animals acquired from external collaborators ensures that the health quality of the colony is maintained at a high level. Some of our strains of mice have impaired immune systems, due to their genetic modification, such as the Athymic nude mouse which have an autosomal recessive nude gene. This allows the growth of human and other non-mouse tissue as the animal's natural immunity is unable to reject the foreign material. These animals demand special care and our

	husbandry and care routines ensure that these animals are maintained in a healthy environment.

Project 2	**The generation, maintenance and breeding of genetically altered mice as a service.**
Key Words	Transgenic, Genetic Alteration, Breeding, Rodents.
Expected duration of the project	5 year(s) 0 months

Purpose of the project (as in ASPA section 5C(3))

Purpose	
Yes	(a) basic research;
	(b) translational or applied research with one of the following aims:
	(c) maintenance of colonies of genetically altered animals
Yes	(i) avoidance, prevention, diagnosis or treatment of disease, ill-health or other abnormality, or their effects, in man, animals or plants;
Yes	(ii) assessment, detection, regulation or modification of physiological conditions in man, animals or plants;

Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed):

This proposed project licence will enable the provision of key services to researchers. The main objectives are;

- 1. To create transgenic mice using pronuclear microinjection techniques.
- 2. Create mice with specific genes that have been “knocked-out” or “knocked-in” using targeted Embryonic Stem cells.
- 3. General breeding and maintenance of Genetically Altered (GA) mice.
- 4. Cryopreservation of embryos and sperm from GA mice.
- 5. Re-derivation of lines using fresh or frozen embryos or those generated by IVF.

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)?

The scientific value of this service. The scientific justification for the animal usage is reflected in; 1. Increased efficiency by using a centralised service. The production of GA mice is technically difficult. We can provide the skills, knowledge and equipment required to efficiently produce transgenic mice with the minimum of animal wastage.

2. Cryopreservation service. Archiving frozen embryos and/or sperm from all modified lines would reduce breeding, and therefore the number of excess animals being produced. It will also ensure a repository is available for future use to guard against the potential loss of a line which would be difficult to replace. 3. Generation of GA animals. This value must be decided on a case-by-case basis and will be provided in the PPL's to which GA mice generated by this service will be transferred, or from which they arose. 4. Maintaining a high animal-health status. Required for many scientific projects by preventing any import of pathogens into the animal facility by the practice of embryo re-derivation.

What types and approximate numbers of animals do you expect to use and over what period of time?

No more than 39,500 rodents will be used over the 5 year project time frame. This is broken down into 30,000 (max) undergoing mild severity and 9,500 (max) undergoing moderate severity procedures.

In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected levels of severity? What will happen to the animals at the end?

The majority of the work will be under a mild severity limit with very little likelihood of any adverse affects to the animals. Any procedures that are carried out under moderate rated protocols have very little chance of adverse affects. However, all surgical procedures will be carried out using sterile techniques to minimise the potential risks of infection to the animal. All animals will be given pain relief immediately after any surgery to avoid pain or potential discomfort. Animals will be either transferred onto other PPL's to be used in further projects, or humanely Killed once they reach their set end point.

Application of the 3Rs

Replacement

In some scientific projects it is not possible to replace the use of rodents. When investigating disease and developmental processes, the complexity of a whole organism cannot always be recapitulated using alternative *in vitro* systems. The tissue, cell and molecular interactions involved in such complex processes cannot be examined in their entirety *in vitro*.

The GA mice generated and bred under this PPL will be investigated for phenotypes and processes that cannot be examined in any other way. All animals bred on this PPL are destined for use in another PPL and the case for that particular model will have been made, and approved, separately in that PPL and/or by that institute's Ethics Committee.

Reduction

The numbers proposed in this programme of work are based on reasonable estimates of generating the required GA mouse lines for the facility over the next 5

years. The advantage of using a centralised transgenic service is that it will decrease the overall number of mice used to generate GA lines. The reasons for this are:

- The availability of highly skilled workers will ensure the lowest number of animals possible will be used.

- Equipment is being used that will maximise the efficiency of transgenic production. Such equipment is costly and could only be purchased by a central service unit.
- Central coordination of animal stock production allows the most efficient use of breeding stock. Excess mice or embryos generated for one project can be used in other transgenic projects.
- Sharing of sterile male mice between projects requiring generation of pseudo pregnant females.
- Making use of our expertise in the areas of sperm and embryo freezing to archive all mutant lines, such that a stock of live mice for each line are not required to ensure lines are not lost.
- The strains that are being used in the facility have been chosen because they are the most efficient for production of transgenic mice.
- New techniques are continually tested and adopted to reduce animal usage significantly. Occasionally excess wild type females are produced, the embryos from which cannot be used at the time for microinjection. These can be cryopreserved, thawed at a later date and cultured to the blastocyst stage when blastocysts are required.

Refinement

The numbers proposed in this programme of work are based on reasonable estimates of generating the required GA mouse lines for the facility over the next 5 years. The advantage of using a centralised transgenic service is that it will decrease the overall number of mice used to generate GA lines. The reasons for this are:

- The availability of highly skilled workers will ensure the lowest number of animals possible will be used.

- Equipment is being used that will maximise the efficiency of transgenic production. Such equipment is costly and could only be purchased by a central service unit.
- Central coordination of animal stock production allows the most efficient use of breeding stock. Excess mice or embryos generated for one project can be used in other transgenic projects.
- Sharing of sterile male mice between projects requiring generation of pseudo pregnant females.
- Making use of our expertise in the areas of sperm and embryo freezing to archive all mutant lines, such that a stock of live mice for each line are not required to ensure lines are not lost.
- The strains that are being used in the facility have been chosen because they are the most efficient for production of transgenic mice.

- New techniques are continually tested and adopted to reduce animal usage significantly. Occasionally excess wild type females are produced, the embryos from which cannot be used at the time for microinjection. These can be cryopreserved, thawed at a later date and cultured to the blastocyst stage when blastocysts are required.

Project 3	**Generation, Breeding and Maintenance of Genetically Altered Rodents**	
Key Words (max. 5 words)	Genetically altered, breeding, DNA, CRISPR	
Expected duration of the project (yrs)	5	
Purpose of the project as in ASPA section 5C(3) (Mark all boxes that apply)	X	Basic research
	X	Translational and applied research
	X	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)	The purpose of this work is to breed rodents with genetic alterations and supply them to work that supports research into serious diseases. Until 1980 mutant models of mice and rats arose mainly by the spontaneous discovery of new variants, but the advent of genetic engineering in the 1980's allowed the artificial manipulation of Deoxyribonucleic Acid (DNA) to create new, carefully designed, genetically altered mice and rats. These have made an enormous contribution to the study of disease and the study of gene function. Research models are becoming more sophisticated and in the future will be engineered to provide researchers with an even better animal model precisely designed to answer questions that will advance the discovery of medicines and provide cures for serious diseases. Although it is possible to identify genes and targets that may cause susceptibility to a certain disease this can only be investigated in an animal model that has	

	been manipulated to study the gene effect. Without animal models it is impossible to determine what effect these changes have on a whole living system. Genetically altered rodents rarely look any different from normal animals and generally their genetic change has no effect on their health and welfare. Genetic alterations are caused in many ways but the two most common are: a) Naturally occurring genetic changes described as mutations and b) Man made changes that add, remove or modify a gene. These are known as transgenics (for animals with added genes) and gene targeted or knockouts animals where a gene function has been removed or altered. c) Some of the protocols e.g. placing embryos into foster mothers require surgery. However any surgical procedure is always done under general anaesthesia with pain relief given. Wherever possible non-surgical methods will be used, i.e Non-Surgical Embryo Transfer (NSET). Genetically altered animals are important in helping to discover what causes disease. For example they may be disease models with a genetic change that mimics a disease like cystic fibrosis in man, or be able to grow implanted human tumours for the study of new anti cancer medicines. Knockout mice allow the function of a single gene to be studied in a particular disease. After being bred, animals will be used for experimental use. This centralised service licence for breeding and supply of mice to scientific projects is administratively efficient, with breeding controlled to produce batches of animals as needed.
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)?	There are several research groups within the establishment who use genetically altered animals to study the genetic basis for disease and abnormalities, such as skeletal development, wound healing, scar prevention, neurological disease and signalling pathways. Without the creation of genetically altered animals this research would not advance (we have over 250 strains at the present moment)

What species and approximate numbers of animals do you expect to use over what period of time?	Mice 200000 Rats 17000
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 majority of strains bred or created with fall into the mild category, ALL strains will be assessed to see if any phenotype is noticed, and amended accordingly. As soon as the scientific objectives have been achieved for each strain, the strain would be frozen down (via freezing embryos or sperm)
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	The generation of genetically altered animals is not possible in vitro, in order to investigate the interaction between individual cells, molecules and growth factors, an animal model is required.
2. Reduction Explain how you will assure the use of minimum numbers of animals	To provide the embryos to implant the embryonic stem cells or DNA, female mice will be given superovulatory drugs to increase the number of ova/embryos produced, this in turn will mean a reduction in the number of female mice used. Once mice have been generated, the offspring produced will be kept to a minimum. Once the strain of mice has been used and the strain is no longer needed, the strain would be cryopreserved (embryos and/or sperm.) The use of new technology Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) means repetition is very rare.
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.	Rodents are used, as they are the least sentient of the species. The techniques used are well established and referenced internationally; data acquired over time has translated well into clinical trials.

Project 4	**Provision of a service for production and maintenance GA animals and antibody production**	
Key Words (max. 5 words)	Service, Genetically altered animals	
Expected duration of the project (yrs)	5	
Purpose of the project as in ASPA section 5C(3) (Mark all boxes that apply)	x	Basic research
		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)	The main aim of this licence is to provide a comprehensive service for the production and maintenance of genetically altered animals. In addition, the licence covers small scale production of serum and antibodies. Genetically altered animals provide complex systems for the study of biological processes. The procedures and protocols that constitute this PPL will result in genetically altered animals being made available for use in a range of project licences involved in medical research. Most of the work is carried out on mice, but rats, frogs and fish are also covered. Protocols used are established and are constantly refined, and the work done under the licence contributes to a huge range of research projects.	
What are the potential benefits likely to derive from this	By providing these services centrally we prevent avoidable animal wastage and ensure experienced	

project (how science could be advanced or humans or animals could benefit from the project)?	personnel do the work. A number of highly trained staff provide the expertise in the production of these animals, meaning that the smallest numbers of animals are used, and that staff are experts in this area, constantly looking to refine practices such as reducing the need for surgery for embryo transfers and using stool samples for genotyping. The service also ensures these genetically altered animals are archived by cryopreserving embryo and sperm to enable their use as a future resource, and to help reduce animal numbers when they are no longer required for active research as well as to enable easy sharing of these animals with scientists across the world. We will also produce sophisticated and refined genetically altered models for research using best practice and novel methods.
What species and approximate numbers of animals do you expect to use over what period of time?	Mice >100,000 Rats ~1000 Fish >40,000 Frogs >10,000 Are anticipated to be used over the course of the 5 year 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?	Most animals used on this licence will be either bred under a mild severity and show no adverse effects or will be used in a mild or moderate surgical procedures resulting in no expected harm due to the nature of the aseptic techniques used and the pre, peri and post operative care they receive. Where animals undergo a moderate procedure we have clear end points and control measures to stop suffering as soon as possible. Animals are killed by a schedule 1 method at the end of procedures or kept alive for breeding.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot	**This service PPL** by its nature requires the use of animals, and will result in GAA being made available for use in most of the PPLs used at the Institute, for which the benefits are clearly described within each

use non-animal alternatives	PPL and will be published via the scientific groups holding these PPLs.
2. Reduction Explain how you will assure the use of minimum numbers of animals	Numbers are kept to a minimum by training staff to high standards, quality control, database tools allowing us to track animals and any adverse effects accurately, through centralising the supply of animals we can reduce wastage and by using specialised staff success rates are high again reducing numbers. The Cryopreservation part of the work is key in reducing the long term animal use.
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.	Rodents, Fish and Frogs are the most frequently used laboratory animals and we have services with best practice established techniques that allow us to provide services for these species to the scientists onsite. **We make every effort to refine our procedures wherever possible, and there is a definite culture of care within the service. Modification of our surgical techniques is happening constantly in line with new developments and best practice to improve welfare. Cryopreservation is encouraged, as a means of reducing the welfare issues involved in animal shipment. We keep up to date of new genetic tools which reduce the severity of phenotypes in animals. Breeding will be kept mild wherever possible, by keeping lines with moderate or severe phenotypes breeding heterozgously wherever possible. New environmental enrichment products are trialled continuously by animal care staff, and there is an active training and development programme for animal care staff ensuring best practice.**

Project 5	**Producing and Maintaining Genetically Altered Rodents**
Key Words	Breeding, Rederivation, Service, Transgenics, Rodents
Expected duration of the project	5 year(s) 0 months

Purpose of the project (as in ASPA section 5C(3))

Purpose	
Yes	(a) basic research;
	(b) translational or applied research with one of the following aims:
Yes	(i) avoidance, prevention, diagnosis or treatment of disease, ill-health or other abnormality, or their effects, in man, animals or plants;
Yes	(ii) assessment, detection, regulation or modification of physiological conditions in man, animals or plants;

Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed):

Genetically altered animals (GAA), particularly rodents, are widely used in biomedical research and are great value for establishing the function of genes and pathways in a variety of biological, physiological and pathological processes. Via this licence, we will provide a specialist technical service for researchers wishing to acquire or create novel GAA for their studies, and protect their colonies against disaster or loss. This licence uses reproductive and transgenic technologies (such as IVF, embryo production, sperm freezing and embryo micromanipulation) to generate and manage GAA animals for users in all departments to minimise animal use and wastage.

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)?

Genetically altered animals are an invaluable tool for understanding disease processes in man and animals and for developing treatments and therapies for them. A number of approaches exist to produce new genetically altered models for disease research. However, these centre largely on the ability to manipulate embryos at early stages of development and then successfully produce offspring from those manipulated offspring. Some methods to achieve this are technically challenging, which can lead to inefficiencies in terms of success when they are used infrequently within individual research groups. The aim of this licence is to offer an expert service for the production and acquisition of animals carrying specific genetic alterations

relevant a broad number of research disciplines. The animals produced or acquired from third parties will be made available at the highest health status to ensure, to the best of our ability, that research outcomes are based on real experimental or treatment effects and not masked or compromised by some underlying pathology resulting from a generally low health status. This approach has advantages in terms of economy and animal welfare, allowing more valuable resources to be deployed into disease research rather than model generation. The work performed under the authority of this licence will be of benefit to a wide number of scientific projects. The animals produced will be used by many different scientists in various disciplines including Cancer Studies, Cardiovascular Sciences, Immunology and Genetics, amongst others. The overall benefit of a centralised service licence to perform these procedures is to reduce duplication and wastage, and to offer an efficient, state-of-the-art service to a wide range of disciplines requiring the use of GA animals for their research.

What types and approximate numbers of animals do you expect to use and over what period of time?

We propose to use mice and rats in this project. Over the total duration of the project, we anticipate using up to 49,500 mice and 4,350 rats.

In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected levels of severity? What will happen to the animals at the end?

The largest proportion of animals accounted for in this project will be produced by natural breeding techniques and are likely to only experience mild phenotypic effects which are not expected to impact significantly on their welfare or wellbeing. Approximately 25% of the animals bred might exhibit more complex symptoms resulting in an anticipated moderate severity assessment. After establishing the genotype of these animals they will be provided to the relevant research groups for further study within the scope of a different Home Office Authority. Those animals used for observation only will be killed by a recognised appropriate method at the end of their period of study, or as required for tissue collection. This project also requires that some animals are subjected to regulated procedures, some of which involve drug administration by hypodermic injection (considered to be of mild severity) and some surgical procedures. In the case where animals undergo surgery, they are not expected to exceed a moderate severity limit. These animals will be killed by a recognised appropriate method at the end of the regulated procedure.

Application of the 3Rs

Replacement

Although many projects use tissue or cell culture systems, unfortunately, these cannot adequately model all of the processes which go on in the human or animal body. As a result, lab-based data is used to help define requirements and expectations for specific research projects, but ultimately it is necessary to establish whether theories based on this data are accurately reproduced in the whole-body.

Reduction

Before producing a new GAA, we will confirm that no other existing model suitable for use by the researcher is readily available, by performing extensive searches of relevant online databases. If a model is available elsewhere we will endeavour to acquire it rather than re-create it. Where no such model exists, we will confirm that any newly created GAA is the most relevant for study before proceeding. Animal breeding programmes will be optimised to avoid the unnecessary production of wasted animals, yet also ensuring that relevant experimental controls are produced where appropriate. Finally, to avoid having to re-create or re-import models, priority will be given to archiving sperm or embryos from each line once established. This avoids the need to maintain live colonies which are not currently in active use by the researcher and provides security against accidental loss.

Refinement

Mice are universally used for studies where genetic alteration is required to understand a disease process. As a result, a large number of reliable and standardised protocols are available for the techniques used in the scope of this licence. The welfare cost to the animals will be minimised by applying the least invasive and most relevant protocols available to achieve the optimum outcome.

Compared to mice, the production of genetically altered rats has not achieved the same degree of success until recent years. Not least because of their larger size, rats are considered useful models for a number of physiological research disciplines therefore increasing success in rat genetic manipulation has resulted in an increased interest in generating models using this species which may be more relevant or produce data more comparable to previous information than a similar mouse model. Once again, the application of proven techniques will minimise the welfare cost to the animals involved. Our choice of species will largely be driven by the requirements of individual researchers.

Project 6	**Breeding and Maintenance of Genetically Altered Rodents**	
Key Words (max. 5 words)	Breeding, Genetically Altered	
Expected duration of the project (yrs)	5	
Purpose of the project as in ASPA section 5C(3) (Mark all boxes that apply)		Basic research
		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)	Genetically altered (GA) rodents are widely used in the fields of biological, medical and veterinary science, and are considered to be of great value in dissecting the function of genes and pathways in physiological and pathological processes. GA animals also provide us with models for many human diseases. The application of GA mice is well established at this establishment to study cancer, parasitic diseases and mechanisms of inflammation. More recently these mice have been used to reveal the development of the lymphoid system and the regulation of autoimmune disease. The data generated from these studies are essential to provide new knowledge at the cellular and molecular level and to validate new prophylactic or therapeutic approaches for diseases. This project aims to breed, maintain and supply GA rodents for our research teams and for those at other establishments working in collaboration with us.	
What are the potential benefits likely to derive from this project (how science could be	A single centralised breeding project, run by individuals with expertise in breeding methodologies and necessary technical skills, together with stringent	

advanced or humans or animals could benefit from the project)?	colony management, will provide an efficient system with subsequent welfare benefits. Effective liaison with research groups will ensure minimal wastage as well as sharing of animal lines and/or tissues whilst maintaining animals of a high health status.
What species and approximate numbers of animals do you expect to use over what period of time?	Mouse breeding, 35,200 over 5 years for use in other Project Licences.
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?	All animals will be housed under specific pathogen-free conditions with autoclaved bedding, food, and water. Animals will be monitored on a daily basis by trained staff as part of the regular husbandry procedures in the facility. The vast majority of animals produced under this licence are not expected to exhibit any harmful phenotype, and will thus be covered by a protocol of mild severity limit. If any adverse effects are observed, advice will be sought from the NACWO and the NVS, and the animals will be humanely killed. For genotyping purposes, a small sample of tissue (usually blood or an ear notch) will be collected. These procedures will result in no more than transient discomfort and no lasting harm. Animals will be transferred to other Project Licences within or external to the establishment for use in research. Some animals may be culled at the end of the breeding cycle and some may be surplus stock. This surplus stock will be minimal, and their tissues will be used for further research whenever possible.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	To date, there are no in vitro assays that adequately model the molecular, cellular and physiological interactions that take place in an intact animal. Nor is it possible to perform studies in which immune/pathological responses to infection of human subjects are manipulated or examined in a controlled manner. Therefore it is necessary to use a whole-animal biological model. The majority of the research work carried out by the groups working with GA animals involves the use of in vitro systems such as cell culture and human tissue assays, but these complement rather than replace the animal studies.
2. Reduction	Unnecessary breeding of genetically altered animals will be avoided by searching cryobanks and

Explain how you will assure the use of minimum numbers of animals	databases. Examples of such resources include: • NC3Rs comprehensive mouse database: http://www.nc3rs.org.uk/category.asp?catID=8 • Jackson Laboratory: http://jaxmice.jax.org/findmice/index.html • Jackson Laboratory Mouse Genome Informatics: http://www.informatics.jax.org/ • Jackson Laboratory Mouse phenonome Database: http://phenome.jax.org/ • Federation of International Mouse Resources: http://www.fimre.org/ Animals will only be bred if a user requirement has been established, and the breeding programme will be subject to regular review to optimise production in line with anticipated demand. Breeding will be optimised, wherever possible, to produce only the genotype required.
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 are universally used for studies involving genetic alterations. They are also the species of choice for the research currently being carried out at the establishment, as much is known about the murine immune system and how it can be manipulated, and reagents to perform such experiments are widely available. Published guidelines for current best practice will be followed to minimise animal suffering. These include: • Refinement and reduction in the production of genetically altered mice: http://la.rsmjournals.com/content/vol37/suppl_1/ • Assessing the welfare of genetically altered mice: see Wells et al (2006) Laboratory Animals 40(2), 111-114 for an overview. • GA passports – the key to consistent animal care: http://www.rspca.org.uk/sciencegroup/researchanimals/reportsandresources

Project 7	**Creation, breeding and maintenance of genetically modified mice**	
Key Words (max. 5 words)	Mice, transgenics, knockout, cryo-preservation, rederivation	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3) (Mark all boxes that apply)	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)	This project will create, breed and maintain mice with genetic alterations and supply them for research into fundamental molecular and cellular functions and disease processes in the fields of biological, medical and veterinary science. For example, this licence supports research on cancer and neurodegenerative diseases, specifically using mice to identify novel treatments.	
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)?	Genetically modified mice (GMM) have made significant contributions to biomedical research. However, the function of many genes is still not known or is not fully understood, either individually or in the ways they interact to produce their intended effects, or how they are dysfunctional in disease. The use of animal models is necessary to determine these processes and to reveal novel treatments for human diseases, which are too complex to be modeled in culture systems. In addition, having a centralized licence to perform the Protocols described facilitates the use of skilled competent staff and facilities for the local dedicated breeding and supply of animals of known health status. This minimises the numbers of animals used and	

	reduces breeding and transport harms.
What species and approximate numbers of animals do you expect to use over what period of time?	Mice, around 50,000 over the 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?	Mice will be used as donors to generate GMM (providing morulas or blastocysts) and recipients females will be used to implant these embryos upon injection of modified stem cells or transgenes. Males may be vasectomised to generate these pseudopregnant recipient mice. GMM will be bred and maintained under this licence only transiently, until mice are transferred to specific experimental licences, Mice bred under this licence do not have any phenotypic consequences, or the phenotype is within the mild severity band. These transgenic mice will then be supplied to individual project licences to perform regulated procedures under their individual Home Office-approved licences. It is important to emphasise that this licence is seeking authority to generate, breed and maintain GM mice, and it is expected that the scientific use of the generated mice will be covered by the requesting scientist's (customer) project licence. The supply and generation of mice under this licence will be justified from a scientific point of view and will have to show a positive harm-benefit balance.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot use non-animal alternatives	The main purpose of this application is to facilitate biomedical research within UCL and beyond, and to complement other National/International efforts aiming to understand human biology using the mouse as a model. In vitro assays cannot adequately model the complete array of molecular, cellular, physiological and behavioural interactions necessary to fully understand how genetic modifications result in normal or abnormal processes. As previously discussed, the scientific use of these GM mice will not be covered by this project licence, but rather, will have to be authorized by the customers' own PPL. The supply and generation of mice under this licence will be justified from a scientific point of view and will have to show a positive harm-benefit balance.
2. Reduction Explain how you will assure the use of minimum numbers	Unnecessary production or import of genetically altered mice will be avoided by searching cryobanks and databases. The supply and generation of mice under this licence will be justified from a scientific point of

of animals	view and will have to show a positive harm-benefit balance. Animals will only be bred if a user requirement has been established, and the breeding programme will be subject to regular review to optimally meet anticipated demand. Once new mouse lines are established in our facilities, as a request of a user, these will be made available for use on other scientific projects for which justification is given in those projects for using the new mouse lines. Breeding will be optimised to produce only the genotype required e.g. Homozygous breeding pairs. Cryopreservation of gametes and embryos to archive lines will avoid wastage from the need to maintain live mice.
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 are universally used for work involving genetic alterations. The standard protocols, methods and reagents have been optimised for this species and there are acknowledged benefits from their use. The Named Veterinary Surgeon and the Named Animal Care & Welfare Officer will oversee the mice of the animals in this project. In general, only mice showing no phenotype or phenotypes within the mild severity band will be bred under this licence. Genetically modified mice may in unexpected and rare circumstances exhibit effects of the disease process that is the focus of the research for which they have been created before such a time that they can be transferred onto the project licence for which they were bred. In this case, we will consult the Home Office inspector as to whether there is adequate scientific justification to keep these animals.alive and the appropriate measures that would need to be taken to minimise the harms they suffer until they are transferred onto the project protocol for which they were originally designed. The methods chosen are all standard for this type of work. Published guidelines for best practice will be followed, including: Refinement and reduction in the production of genetically modified mice; Laboratory Animals Vol 37, Supp 1 July 2003. GM mouse welfare assessment working group http://cbctraining.ncl.ac.ukleM-EU5/story. Html Assessing the welfare of genetically altered mice. Wells et al (2006) Laboratoiy Animals 40(2), 111-114 Laboratory Animal Science Association Good Practice Guidelines Series 1/Issue 1) October 1998. Administration of Substances (Rat, Mouse, Guinea Pig, Rabbit) and Collection of Blood Samples (Rat, Mouse, Guinea Pin, Rabbit).

Project 8	**Cryopreservation of Mouse and Rat Embryos**	
Key Words (max. 5 words)	Cryopreservation Mouse and Rat Embryos	
Expected duration of the project (yrs)	5 years	
Purpose of the project as in ASPA section 5C(3) (Mark all boxes that apply)	X	Basic research
		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)	The main purpose of this licence is to ensure reduction in animals held overall for studies. A large number of genetically altered (GA) mouse and rat strains are required for research at the institution. The overall objective of this licence is to continue to provide a centralised cryopreservation service for GA mouse and rat embryos in our establishment. Typically small numbers of animals up to 10 females are required to be superovulated to produce up to 200 cryopreserved embryos, which will secure each transgenic line. These 200 embryos can be held in storage to enable the line to be revived at any time on a number of occasions, if required. In our experience for re-implantation only small numbers of mice, typically 2 females, are required for this procedure when a transgenic line is required to be resuscitated and viable mice generated. Our staff have considerable experience and expertise to ensure that breeding programmes are	

	co-ordinated and to ensure optimal animal husbandry and minimal animals being used. All of the methods used on this licence are established standard validated techniques with a proven high success rate; however, we constantly seek new methods of refinement of the procedures used e.g. the use of sterile males mice rather than vasectomised animals in mice. There is no non-animal alternative available.
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)?	Many of the researchers themselves may not have the technical skills to carry out cryopreservation and re-implantation techniques in the mouse and rats, therefore a cryopreservation service was established. Since then, many users have taken advantage of this service, which is administratively efficient, with procedures controlled to minimise the use of animals. Cryopreservation reduces the number of transgenic or mutant mouse lines required to be maintained by "ticking over" breeding because they are not required for current studies and therefore overall limits any chance of over production of any surplus animals. Cryopreservation also provides an insurance against potential loss of valuable lines.
What species and approximate numbers of animals do you expect to use over what period of time?	Rat and mice. Accurate numbers are difficult to predict as it depends on demand on the service however over 5 years it is very unlikely to exceed 3000.
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?	Mild protocols 1-3 . Moderate on procedure 4. Schedule 1 killed at end of procedure or transferred to licences with authority.
Application of the 3Rs	
1. Replacement State why you need to use animals and why you cannot	No alternative to the use of live animals for the production of ova, sperm or embryos or in the creation of pseudo pregnant female animals.

use non-animal alternatives	
2. Reduction Explain how you will assure the use of minimum numbers of animals	Female animals are superovulated in small numbers and the number of embryos cryopreserved reviewed before further animals are superovulated. Once adequate numbers of embryos are harvested no further superovulation will be carried out. Numbers of stud males are based on previous experience to allow rotation of the males and prevent overuse. Numbers of pseudo-pregnant females are calculated from previous experience to prevent wastage of embryos and females.
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 vasectomy process is refined by the use of naturally sterile male mice when commercially available rather than surgically prepared vasectomised animals for mating to produce pseudo-pregnant female mice. When sterile males are not available surgical prepared vasectomised animals will be used. The scrotal approach to vasectomy rather than a more invasive laparotomy is carried out. Pain will be controlled peri-operatively by general anaesthesia and analgesia on the advice of the Named Veterinary Surgeon. This is a continually developing field and new and better refined techniques such as non-surgical implantation of embryos will be explored as they become available and introduced once established as a refinement.

Project 9	Breeding genetically altered mice and rederivation
Key Words	Breeding genetically altered mice and rederivation
Expected duration of the project	5 year(s) 0 months

Purpose of the project (as in ASPA section 5C(3))

Purpose

Yes (a) basic research;

(b) translational or applied research with one of the following aims:

Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed):

Mice are bred and transferred to authorised projects. Animals may be obtained from other establishments. They will be quarantined and health screened before being transferred to the main animal facility. If an infection is present, the embryos of mice will be taken and transferred to foster mothers free of disease. This process is called re-derivation. In some cases the animals are no longer required but are potentially very valuable to scientists in the future, therefore the embryos of these animals will be frozen for future use.

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)?

A centralised breeding programme for a range of research projects ensure the best husbandry conditions and efficient record keeping. It makes it possible to minimise waste and to ensure that no more animals are produced than are required for the various research projects. We are also able to maintain the genetic stability of the strains that we breed by using the best breeding strategies. This ensures that all of the mice stay the same genetically, which is beneficial to the science. The animals are then supplied to scientific projects within the institution, which have undergone ethical and peer review

What types and approximate numbers of animals do you expect to use and over what period of time?

It is anticipated that the number of mice bred or used each year under the authority of this licence will be no more than 3000

In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected levels of severity? What will happen to the animals at the end?

For most of the animals the severity of the procedures will be mild. The animals with possible adverse phenotype will be closely monitored for any health and welfare

issues. Vasectomy and the surgical procedure by which mouse embryos are implanted in pseudo-pregnant recipient animals will result in a moderate level of pain and possible adverse effects include wound infection or an adverse reaction to anaesthesia. These will be minimised by using aseptic surgical technique, analgesia and by closely monitoring any animals that undergo surgery. Any animals which cannot be used for scientific research will be humanely killed using a Home Office approved (Schedule 1)method and tissues provided to researchers for ex-vivo projects where possible.

Application of the 3Rs

Replacement

Where possible, advancements in medical research forward using cell cultures and other in vitro methods, without having the needs to use live animals. However, there are still many areas where the study of disease and the quest for treatments and remedies can only be pursued effectively in scientific studies involving the use of living animals.

However, the ultimate justification for using animals lies with the end users' project licence. This will have been subject to ethical, peer and Home Office approval before being granted.

Reduction

The use of computer database system for breeding records maximises the efficiency and regular discussions with the end users ensure the numbers of animals bred match the demand and do not exceed requirements. The use of superovulation significantly increase the numbers of embryos which can be harvested from each mouse and therefore means that fewer animals are required overall.

Refinement

The justification of using a particular strain of mice will lie with the end users' project licence. We will always use best practice when giving injections or carrying out surgical techniques. Analgesia will be provided prior to surgery. Non-surgical methods of embryo transfer will be evaluated and if these methods prove superior to surgical methods they may be adopted as standard Appropriate specialised accommodation is provided. All strains are bred in a fashion to avoid infection. Routine health checks are carried out to monitor welfare. The animals are bred by a dedicated team of trained staff with experience in the care of such animals. Environmental enrichment, including nesting material is provided to accommodate natural behaviour of the mice.

Project 10	Breeding and maintenance of GA mice.
Key Words	Breeding, Maintenance, Genetically, Altered, Mice
Expected duration of the project	5 year(s) 0 months

Purpose of the project (as in ASPA section 5C(3))

Purpose

Yes (a) basic research;

Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed):

The aims and objectives of the project are to breed and maintain established colonies of Genetically Altered (GA) mice. These mice will be used for scientific research, which may require separate Project licence authority. The project will allow us to acquire new lines of GA mice that are of interest to our researchers and it will allow us to cryopreserve embryo and sperm from GA mice when they are not being actively studied. It will also allow us to rederive colonies to improve animal health and welfare.

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)?

This project is a continuation of a service licence to provide breeding and maintenance of genetically altered mice. A team of technicians with expertise and experience will care for these mice. Researchers will be provided with mice of a high quality that will allow for greater reproducibility in their results. The ability to cryopreserve embryos and sperm will lead to a reduction in the numbers of mice being bred. Rederivation will improve animal health and welfare and ensure that we provide high quality mice for our researchers Results from studies undertaken on mice bred under this licence will be published in peer reviewed journals and lead to increase in the knowledge and understanding of the models being studied.

What types and approximate numbers of animals do you expect to use and over what period of time?

Mice, all stages, 48,130 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 levels of severity? What will happen to the animals at the end?

There are no expected adverse effects from breeding of GA mice. Females of an appropriate size will be used and over vigorous males will be replaced. To identify the mice and to check the genetic make up, a small tissue sample (normally from the ear) may be taken. This may cause transient pain, but trained and experienced people take the sample. Some female mice (less than 1% of the total animals requested) may be injected with small volumes hormones to increase the number of embryos they can produce, a procedure known as superovulation. The needle insertion may cause transient pain. Some mice (less than 1% of the total animals requested) may be anaesthetised for surgical procedures, this may by injection or by inhalation anaesthetic. This will be for relatively short periods of time. After being anaesthetised, one of two surgical procedures may be performed: either vasectomy in males or embryo transfer in females. For the vasectomy, a small cut is made in the scrotal wall and a small piece of the vas deferens cut away. The hole in the scrotal sac is then repaired. The mice are given analgesia before and after the surgery. For embryo transfer, a small cut is made on the back of the mouse, through the body wall. The uterus or oviduct is exposed and a small hole is made in the uterus or oviduct, embryos held in a very fine tipped pipette are then transferred into the uterus or oviduct. The incision is repaired and the mouse allowed to recover. The mice are given analgesia before and after the surgery. Animals are expected to make a rapid recovery after the anaesthetic. Mice with genetic alterations are affected in different ways depending on the gene/s affected, many look and behave as normal mice. In this project, some of the GA mice born may have balance problems making their walking unsteady. Other strains used in Alzheimer's research, may lose the structure or function of the cells in the brain, which may cause changes in their normal behaviour over time. Sometimes the mutation makes the mice smaller than their normal litter mates. All of these mice are monitored closely. To ensure that these animals can reach the food and water in their cage they will be provided with wet mash The animals may be transferred to another authorised project, kept alive at the establishment or humanely culled. Trained and experienced personnel will carry out the procedures.

Application of the 3Rs

Replacement

Animals are required as although many research projects involve the use of in-vitro systems such as cell culture, human tissue assays and computer modelling, *these* cannot yet adequately model all aspects of the complex biological process involved.

The chosen species for this project is the mouse. Mice are biologically very similar to humans and mice can be genetically manipulated to mimic many human diseases.

Reduction

The project licence holder and animal technicians have a good working knowledge of the colonies being bred and a good working relationship with the researchers. The breeding colonies will be managed efficiently to meet the research needs.

Each request for new GA mice will be reviewed by a committee and we will review the breeding colonies regularly and meet the research group to discuss their requirements.

Refinement

Mice are the model of choice as their genetic, biological and behavioural characteristics closely resemble those of humans.

The mice will be cared for by animal technicians and veterinary staff who have experience of the husbandry and welfare relevant to their needs.

Experienced and competent personal licence holders will conduct the procedures and veterinary advice will be taken in respect to the use of anaesthesia, analgesia and aseptic technique.

Project 11	Breeding and Production of Genetically Altered Mice
Key Words	Genetic alteration, mouse, breeding
Expected duration of the project	5 year(s) 0 months

Purpose of the project (as in ASPA section 5C(3))

Purpose	
Yes	(a) basic research;
	(b) translational or applied research with one of the following aims:
Yes	(i) avoidance, prevention, diagnosis or treatment of disease, ill-health or other abnormality, or their effects, in man, animals or plants;
Yes	(ii) assessment, detection, regulation or modification of physiological conditions in man, animals or plants;

Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed):

This project will breed transgenic mice for use in biomedical research. These animals are needed to investigate a range of human diseases and also to help understand the influence of different genetic factors in human development.

The mice bred on this project will be used on a range of different projects that only require small numbers of animals. Centralising our breeding of small groups of mice ensures they are cared for by experienced technicians, who are also trained in assessing possible adverse effects of the genetic alterations. We also ensure that the minimum number of mice are bred by carefully monitoring colony size.

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)?

The types of research project undertaken using these animals include investigations of the mechanisms of several inherited human diseases, development of novel methods of treating cancer, the mechanisms underlying some forms of male infertility, and a range of studies investigating early human development.

What types and approximate numbers of animals do you expect to use and over what period of time?

Mice will be used, and it is expected that up to 5500 mice will be used 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 levels of severity? What will happen to the animals at the end?

A few animals (c.200) will be used to establish new breeding colonies at our facility. These animals will undergo surgical implantation of embryos that have been genetically altered, and these animals will give birth to transgenic offspring that are used to start a breeding colony. In order to prepare the mice for implantation of the eggs, they are mated with male mice who have been made sterile by vasectomising them (a procedure also undertaken as part of this project, to approximately 100 mice). Alternatively, adult mice are imported, and these are bred to produce more transgenic mice. To check their genetic make-up, small tissue samples may be taken, for example a small piece of ear or tail. Pain-killing drugs or anaesthetics are given whenever these are likely to be needed, so that the minimum of pain and distress is caused. Although all of the mice bred for these projects have genetic alterations, most of them show very minor or no adverse effects. Other types of mice are normal when they carry only one copy of the altered gene, but would be abnormal if they have two copies of the altered gene. To reduce the number of animals that are produced with adverse effects (such as abnormal development of particular tissues or organs), many studies are carried out on tissues obtained from animals humanely killed at an early stage of development.

Application of the 3Rs

Replacement

Many of the research groups working on mice bred on this project also undertake studies in humans, in human and mouse tissue culture, and in other animal models such as fish, and only use mice when it is absolutely necessary for theparticular research that they are undertaking.

Some work, especially that which involved studying the development of multiple organ systems requires some work in whole organisms. This is because the complex interactions between different organ systems cannot yet be studies fully in isolated cells and tissues grown in a dish in the laboratory.

Reduction

We will minimise animal use by only performing work in whole animals when alternative are unsuitable, or when work cannot be conducted directly in people. We will minimise the numbers of mice bred by careful colony management. We will aim to make tissues available from any animals bred to other research groups either directly, or by participating in national and international schemes for sharing such resources. Where specific types of mice are readily available from academic or commercial sources, mice will be acquired for each study, to avoid maintaining a breeding colony.

Refinement

Mice are used for these projects, because the genetic make-up of the mouse has been successfully decoded, and all of its genes identified. This information is now being used to determine the function of these genes, and in particular how they interact in a whole animal. The welfare costs of the work are minimized in two main ways. When animals undergo surgery (for vasectomy or embryo transfer) pain is prevented by use of analgesics, and distress is minimized by high standards of perioperative care. When genetically altered mice are bred, then they are monitored for any adverse effects of the genetic alteration. If there is any harmful effect, then carefully defined criteria are established to limit these effects. This usually involves humanely killing the animals before their health is compromised, but most of the animals bred on the project remain clinically normal throughout their life-span.
When a new type of genetically altered mouse is to be imported, details of the anticipated effects of the genetic modification will be obtained from the supplier and this will inform the initial decisions in relation to breeding and care of the mice. When possible, “mouse passport” data that contains more specific husbandry advice will be sought. During establishment of the initial breeding colony, litter size, number successfully weaned, and any specific adverse effects will be documented by regular (daily) examination of the animals. Husbandry modifications (eg use of soft diet, later weaning dates for smaller juveniles, additional bedding etc) will be adopted as required. If the genetic alteration could lead to a reduced resistance to infections, we would change the way we house the mice to reduce the risk of them being exposed to disease agents.