



TOP TIPS FOR MANAGING
MOUSE BREEDING COLONIES

UQ RESEARCH AND INNOVATION



THE UNIVERSITY
OF QUEENSLAND
AUSTRALIA



Create change



UQ RESEARCH AND INNOVATION

UQ's innovations have made, and continue to make, life-changing advances. The University is committed to maintaining the highest standards of ethical decision-making, animal husbandry and care as a contribution to achieving quality research and teaching outcomes.

For research that involves the study of animals in the wild or in agriculture, or the use of animal models of human disease, the University's Animal Ethics Committees review proposals to ensure that animal welfare is fully considered before the commencement of any activities. Similarly, the Committees must consider teaching activities that directly involve animals, such as veterinary science or marine biology.

As part of the Top Tips Series produced by UQ Research and Innovation (UQR&I), this brochure provides advice to researchers to assist with management of laboratory animal breeding colonies at The University of Queensland, including ethics applications, managing the colony and preparing Mandatory Annual Reports



CONTENTS

	Introduction	03
1.	The breeding colony application	03
2.	Before you begin	03
3.	Justification of the breeding strategy	04
4.	Example 1: Justifying animal numbers	04
5.	Example 2: Question 16 - Entering breeders	05
6.	Example 3: Question 18a - Entering production numbers	05
7.	Example 4: Question 18b - Entering cull numbers	05
8.	Example 5: Breeding Animal Ethics Approval Certificate	06
9.	Genotrack	07
10.	Genotrack reports: Definitions	07
11.	Mandatory Annual Report of animal use	09
12.	Example 6: Mandatory Annual Report	09
13.	Modification to an approval for a breeding colony	11
14.	Unexpected adverse events in breeding colonies	12
15.	Expected welfare risks in a breeding colony	12
16.	Events to report to the Animal Ethics Committee	12

INTRODUCTION

Breeding colonies of laboratory animals must be professionally managed to avoid or minimise the production of excess animals. The University of Queensland (UQ) requires that animal ethics approval is obtained prior to establishing a breeding colony. Both breeding and research colonies are managed with the help of UQ Biological Resources (UQBR) and by the use of Genotrack. Genotrack software has been implemented to meet the record keeping requirements for breeding and research colonies at UQ, and to assist staff in providing a high standard of breeding colony management.

01

THE BREEDING COLONY APPLICATION

When you apply for Animal Ethics Committee approval for a UQ laboratory animal breeding colony you must first complete and submit the 'Breeding Colony Application Form'.

Through completion of the Breeding Colony Application Form you will advise the Animal Ethics Committee of the following:

- The reason for breeding the animals.
- The breeding strategy to produce the animals needed for research or teaching use, while not overproducing animals that will not be used.
- Phenotypes associated with genotypes that may introduce a welfare risk to the animals.
- If appropriate, methods that will be put in place to minimise the welfare risk.
- The people responsible for managing the day to day care of animals in the breeding and research colonies.

Download the Breeding Colony Application Form using the link found on the [Forms and Templates](#) page of the Animal Ethics @ UQ website.

02

BEFORE YOU BEGIN

The strain names that you will be asked to provide in the Breeding Colony Application Form will be used to populate:

- the Animal Ethics Approval Certificate for the breeding colony
- the form fields for your Mandatory Annual Report
- the Genotrack entries for the breeding colony

Consistent naming of the animal strain between different Animal Ethics Approval Certificates (breeding, research and teaching) will simplify long-term colony management and animal reporting.

03

JUSTIFICATION OF THE BREEDING STRATEGY

A justification for a breeding colony should include:

- The reason for maintaining a breeding colony of the strains that will be listed in the application form.
- If a strain is already part of an existing breeding colony at UQ, the justification as to why it is necessary to establish a second breeding colony.
- The number of animals needed for the research or teaching experiment. This is the reason for establishing the breeding colony and the breeding strategy should be based upon this number.
- The proportion of animals produced that will be used for experiments.
- Information regarding the reproductive performance of the mouse strain.

Information related to mouse reproductive performance can be obtained from online mouse resources, or you may use statistics from your own breeding colonies. If you are unsure, the Officer in Charge of the UQBR animal facility in which your animals will be housed, will be able to help you.

[The Jackson Laboratory](#) website is a valuable source of mouse strain information. You may download the free Handbook of Genetically Standardized Mice that contains tables describing the reproductive performance of several mouse strains.

As an example, for C57BL/6J mice the following information is provided:

Number of pups weaned per female (mean)	29.5
Mean maternal age (days); birth of first litter	67
Maternal age; last litter weaned	8.3 months
Number of litters per female (mean)	5.4
Litter size at birth (mean)	5.9
Litter size at weaning (mean)	5.5

Image: The Jackson Laboratory are mouse breeding colony experts. Your own breeding colony might not be as efficient.

04

EXAMPLE 1: JUSTIFYING ANIMAL NUMBERS

Question 17. Justify the number of breeding animals maintained or to be maintained:

Animals required for experiments: Work being conducted under AEC approval number 123/15 requires 300 homozygous animals that we will use over three years. Therefore, we need to produce 100 homozygous animals per year.

Breeding strategy: Heterozygous x heterozygous mating pairs

Expected genotypes: It is expected that one quarter of the offspring will be homozygous

Number of breeding animals: We will maintain seven breeding pairs at any time and breeders will be replaced each six months.

The expected production numbers per year are:

- 14 females x 5.4 litters per female* x 5.5 pups per litter* = 416 animals produced.
- Of the 416 animals produced, 104 are expected to be homozygous.

* Expected numbers for C57BL/6J mice are from the Handbook of Genetically Standardize Mice (Jackson Laboratories)



05

EXAMPLE 2: ENTERING BREEDERS QUESTION 16.

Number of breeders required per year (please include replacement breeders in this number).

Example 1 justified the number of breeding animals required to produce the number of animals needed for experiments. Question 16 of the Breeding Colony Application Form asks for the number of breeding animals per year. Table 1 is an example showing entry of information into Question 16 of the Breeding Colony Application Form based on the justification in Example 1. Entries in Question 16 will be used to populate the Animal Ethics Approval Certificate for the breeding colony.

Table 1: Entering breeding animals in the Breeding Colony Application

Species (common name)	Strain	Gender	Class of animals	No. of breeders
Mouse	Genotrack colony name	Mixed	GMO	28

If the breeding strategy requires that transgenic mice be mated with C57BL/6 mice, then two rows will need to be completed to request 14 transgenic mice, and 14 C57BL/6 inbred mice. This distinction will become important when you order animals for your colony. Table 2 provides an example of how to enter this information into Question 16 of the Breeding Colony Application Form.

There is no need to include the ages of animals being requested on a breeding application form. It is assumed that breeding animals are adults and animals that are not needed could be culled prior to weaning or after weaning depending upon the circumstances.

Table 2: Entering multiple strains for a breeding strategy in the Breeding Colony Application

Species (common name)	Strain	Gender	Class of animals	No. of breeders
Mouse	Genotrack colony name	Female	GMO	14
Mouse	C57BL/6	Male	Inbred	14

06

EXAMPLE 3: ENTERING PRODUCTION NUMBERS

Question 18a. How many animals do you hope to produce from this colony (production numbers) per year?

Example 1 included a calculation for the expected number of animals that will be produced. This information should be placed into the table at Question 18a of the Breeding Colony Application Form. The numbers for Example 1 have been entered into Table 3.

Table 3: Entering production numbers in the Breeding Colony Application

Species (common name)	Strain	Gender	Class of animals	No. produced
Mouse	Genotrack colony name	Mixed	GMO	416

07

EXAMPLE 4: ENTERING CULL NUMBERS

Question 18b. What is the estimated number of animals to be culled from this colony per year?

Culls in the context of the Breeding Application Form are any animals that are not expected to be used for experiments or as replacement breeders. From Example 1 the cull animals are calculated as follows:

Total produced	416
Minus animals used in experiments	100
Minus animals used as breeders	28
Total culls	288

This information should be placed into the table at Question 18b of the Breeding Colony Application Form. The numbers for Example 1 have been entered into Table 4 and 5 below. This table at Question 16 will be used to populate the Animal Ethics Approval Certificate for the breeding colony.

Table 4: Entering cull numbers in the Breeding Colony Application if all breeding animals were sourced from the breeding colony

Species (common name)	Strain	Gender	Class of animals	No. of culls
Mouse	Genotrack colony name	Mixed	GMO	288

Table 5: Entering cull numbers in the Breeding Colony Application if animals from the breeding colony were being bred with C57BL/6 inbred mice

Species (common name)	Strain	Gender	Class of animals	No. of culls
Mouse	Genotrack colony name	Mixed	GMO	302

08 EXAMPLE 5: BREEDING ANIMAL ETHICS APPROVAL CERTIFICATE

The information provided in Questions 16 and 18 of a Breeding Colony Application Form are used to populate the Animal Ethics Approval Certificate for the breeding colony. Table 5 demonstrates the information that would populate the Animal Ethics Approval Certificate that would be issued if the Examples above were approved by the Animal Ethics Committee. Note that the certificate lists 'breed' and 'cull' animals separately for each approved strain. The numbers provided in Table 6 assume that the approval period was only one year. For three year approvals the number of animals that were justified for use per year in the application, would be multiplied by three when the certificate is created.

Table 6: A sample of an Animal Ethics Approval Certificate for a breeding colony

Subspecies	Strain	Class	Gender	Approved
Mice - GMO	Genotrack Colony Name (breed)	Adults	Mixed	14
Mice - GMO	Genotrack Colony Name (cull)	Adults	Mixed	302
Mice - Inbred	C57BL/6 (breed)	Adults	Mixed	14

When submitting research or teaching applications to use animals from your breeding colony, make sure that the strain name is the same as shown on your breeding colony certificate. This will enable easy generation of Genotrack Reports for annual reporting of animal use under both your breeding and research or teaching Animal Ethics Approval Certificates.

You should not assume that Animal Ethics Unit staff have training in genetics or genetic nomenclature. If you write Gene^{-/-} on one application and Gene KO on another, those will be the approved strain names on your certificates. That might create confusion when you receive your Genotrack records and produce reports for preparing Mandatory Annual Reports. Animal Ethics Unit staff are unable to interpret or change what you have written in your application.



Genotrack is an animal colony management software package that has been implemented at UQ to maintain records of laboratory animals housed in UQBR animal facilities. Every animal housed in a UQBR animal facility is recorded in Genotrack. Once laboratory animals have been approved for use by the Animal Ethics Committee (breeding, research or teaching application), the information from the Animal Ethics Approval Certificate is entered into Genotrack by UQ Biological Resources staff. The Genotrack records will include:

- the Animal Ethics Approval number
- the strain name as written on the Animal Ethics Approval Certificate
- the number of animals approved for use
- the participants who are approved to work with animals under the approval number
- in some cases, the facilities that are approved to house the animals
- the approval expiry date

Managing your colony

Genotrack is a valuable resource for UQ's research and teaching staff who manage laboratory animal breeding colonies. The following are some useful functions of Genotrack:

- Review the live animals in your colony without the need to visit an animal facility
- Identify animals you wish to reserve for experimental use.
- Request that UQBR staff cull excess or unneeded animals
- Determine whether the colony is producing the expected number of pups per litter
- Determine whether female mice have the expected number of litters.

Creating reports

You can generate reports of animal use recorded by Genotrack colonies. Genotrack can help you:

- Determine how many animals are remaining for use under an Animal Ethics Approval Certificate
- Determine how many animals were used during a defined time period. This feature will be of use when completing the Mandatory Annual Report of animal use.

It is recommended that participants who manage breeding colonies obtain training in the use of Genotrack. It is also recommended that they meet regularly with the UQBR staff that work in the rooms in which their animals are housed.

Contact [UQ Biological Resources](#) to arrange a personal or group training session.

GENOTRACK REPORTS: DEFINITIONS

Generating a Genotrack report of animals used in a reporting period is a helpful way to collect the information that will need to be entered into the Mandatory Annual Report for a breeding colony. A Genotrack report will contain a list of animals in a variety of 'states' that include:

Alive (available)

Animals that were produced in the breeding colony and are still alive at the time that the report is generated. These animals would not yet have been transferred to a research or teaching approval, and would not have been used as breeders in the breeding colony.

Dead (culled)

Animals produced in the breeding colony and were dead at the time that the report is generated. These animals were not transferred to a research or teaching approval, and were not used as breeders in the breeding colony.

Dead (found dead) or Dead (found dead) juvenile

Genotrack entries were created when these animals were born. They died while they were in the colony. This category includes animals that were born and were subsequently found dead.

Dead (retired breeder)

These animals were breeders in the colony during the reporting period, but they were retired and euthanased or died before the Genotrack report was created.

Reserved (mating)

These animals were alive during the reporting period and are either the current, live, breeding animals in the colony, ex-breeders, or have been reserved as future breeders.



11

MANDATORY ANNUAL REPORT OF ANIMAL USE

A Mandatory Annual Report of animal use is required for each current Animal Ethics Approval Certificate and reports the number of animals used during each reporting period (January 1 - December 31 of the report year). The animals that should be reported in Mandatory Annual Report for a Breeding Colony Approval are:

Breed

- Female and male animals that are placed in the same cage with the aim of producing offspring. Breeders can be animals that were born into the same colony, an ordered inbred strain, such as C57BL/6, or an animal from another UQ breeding colony.
- Males and females that were placed in a cage to mate, but did not produce any litters.

Cull

- Animals produced in the breeding colony that were euthanased during the reporting period, or
- Animals produced in the breeding colony, that died in the colony. This could be animals that were cannibalised or died due to unknown causes.
- Culls DO NOT include retired breeders that were euthanased during the reporting period. Animals used as breeders will be reported as 'Breed' animals only.

DO NOT report animals that were transferred for use under a research or teaching Animal Ethics Approval Certificate in the Mandatory Annual Report for the breeding colony. These animals should only be reported under the approval number for the research and teaching activity.

DO NOT report animals whose fate remains unknown (with a state of 'Alive (available)'). These animals have not been 'used' yet and might become breeders or culls in the breeding colony in the following year, or be transferred to an Animal Ethics Committee approved

experiment in the following year.

Reporting of breeding animals that are breeding across two reporting periods

An animal that is a breeder on 31 December of one year, and continues as a breeder on 1 January in the following year, must be reported in the Mandatory Annual Report for both years. This is a reporting requirement of the Queensland Animal Care and Protection Act (2001)

As an example, a breeding box with 1 male and 1 female breeder was established on 1 December, 2015 and dismantled on 1 February, 2016.

The two animals will be reported as used animals in the Mandatory Annual Reports for both 2015 and 2016.

12

EXAMPLE 6: MANDATORY ANNUAL REPORT

Genotrack Report for January 1 - December 31, 2015

State: Alive (Available)	50
State: Dead (Culled)	50
State: Dead (Found Dead)	10
State: Dead (Retired Breeder)	10
State: Reserved (Mating)	10

2015 Mandatory Annual Report - Cull animals

Dead (culled)	50
Dead (found dead)	10
<hr/>	
Total to report	60

2015 Mandatory Annual Report - Breed animals

Reserved (Mating)	10
Dead (Retired Breeder)	10
<hr/>	
Total to report	20

2016 Mandatory Annual Report

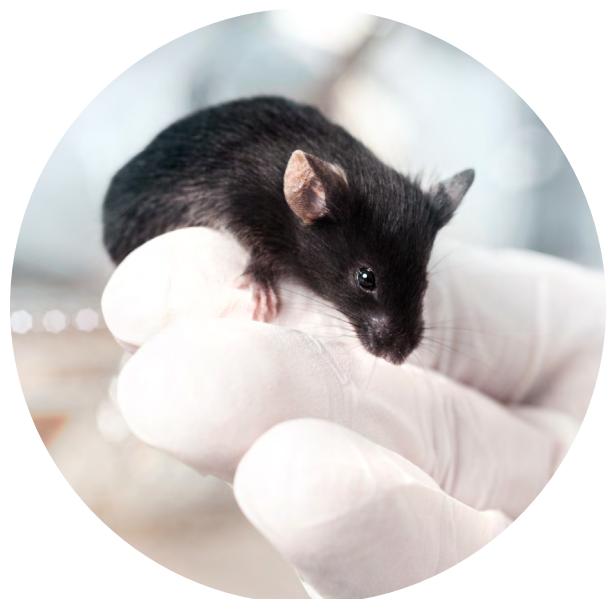
Reserved (Mating)	10
-------------------	----

These animals were breeding in 2015 and continued as breeders into 2016. They should be reported in the Mandatory Annual Reports for both years. Note that the Genotrack report generated at the end of 2016 will list these animals as 'Dead (Retired breeder)'.

Available Stock 50 animals

The reporting of these animals in 2016 will depend upon how they are used. They will fall into one of the following categories:

- Animals transferred to an experiment will be reported under the research or teaching approval number
- Animals that become breeding animals in the breeding colony will be reported as a breeder
- Animals that are culled or are found dead in the colony will be reported as culls



MODIFICATION TO AN APPROVAL FOR A BREEDING COLONY

There are several instances where you may wish to request an amendment to the Animal Ethics Approval for your breeding colony.

Adding additional animals of an approved strain

When applying for animal numbers on a Breeding Colony Application Form, animal numbers are justified based on expected breeding. Researchers should monitor and review their breeding colony regularly to ensure that it is producing as expected.

In Example 1, the justification was based on one quarter of the pups containing the correct genotype for experiments. If the observed number is less than expected then the breeding strategy will need to be amended.

The justification for the change to the approved procedures might be:

The breeding strategy justified in the original application was based upon the expectation that one quarter of animals produced would be homozygous and that the average litter size at weaning was 5.4.

During the first 6 months of breeding, only one fifth of animals were homozygous and the average litter size at weaning was observed to be 4.6.

It is necessary to amend the strategy to obtain the 100 homozygous animals per year that are required for experiments.

Breeding strategy: Heterozygous x heterozygous mating pairs

Number of breeding animals: We will increase the number of breeding pairs to maintain the colony from 7 breeding pairs at any time to 11. Breeders will be replaced every 6 months.

The expected production numbers per year are:

- 22 females x 5.4 litters per female x 4.6 pups per litter = 546 animals produced.

- Of the 546 animals produced, 109 are expected to be homozygous.

	Approved animals (per year)	Revised animals (per year)	Additional required (per year)
Genotrack Colony Name (breed)	14	22	8
Genotrack Colony Name (cull)	302	424 (546-100-22)	122
C57BL/6 (breed)	14	22	8

The Animal Ethics Unit staff will multiply the 'additional required (per year)' by the time remaining on the certificate (e.g. 1.5 years) to ascertain the number of animals that will be added to the approved strain on the Animal Ethics Approval Certificate.

Add an additional strain to an Animal Ethics Approval Certificate for a breeding colony

To add an additional strain to an Animal Ethics Approval Certificate for a breeding colony, the Animal Ethics Committee must be provided with the same information for each strain that was requested in the Breeding Colony Application Form.

- The reason for establishing a breeding colony
- The breeding strategy to produce the animals needed for research or teaching use, while not overproducing animals that will not be used. Include:
 - the number of breeding animals required per year
 - the expected number of animals that will be produced each year
 - the expected number of animals that will be culled per year
- Phenotypes associated with genotypes that may introduce a welfare risk to the animals.

Provide a clear summary of the additional animals required that will appear on the amended Animal Ethics Approval Certificate. For example, if the same breeding strategy as Example 1 was being used:

Summary of additional animals requested per year:

Genotrack Colony Name 2 (breed), GMO, mixed sex	14
Genotrack Colony Name 2 (cull), GMO, mixed sex	302
C57BL/6, inbred, mixed sex	14

14

UNEXPECTED ADVERSE EVENTS IN BREEDING COLONIES

The Australian code for the care and use of animals for scientific purposes defines an Unexpected Adverse Event as:

An event that may have a negative impact on the wellbeing of animals and was not foreshadowed in the approved project or activity.

An unexpected adverse event may result from different causes, including but not limited to:

- death of an animal, or group of animals, that was not expected (e.g. during surgery or anaesthesia, or after a procedure or treatment)
- adverse effects following a procedure or treatment that were not expected
- adverse effects in a larger number of animals than predicted during the planning of the project or activity, based on the number of animals actually used, not the number approved for the study
- a greater level of pain or distress than was predicted during the planning of the project or activity
- power failures, inclement weather, emergency situations or other factors external to the project or activity that have a negative impact on the welfare of the animals.

15

WHAT IS EXPECTED IN A BREEDING COLONY

Expected adverse events for animals in a laboratory animal breeding colony will depend upon the species and strain being used. The majority of breeding colonies at UQ are for genetically modified mice. There is a wealth of knowledge of the characteristics and reproductive performance of the commonly used strains of mice for scientific research are readily available.

It is recommended that researchers consider this information when selecting which strain to use for their research, and in devising appropriate breeding

strategies. For example, UQ has a large number of breeding colonies of genetically modified mice which are, for the most part, bred on a C57BL/6 background. A great resource for information for this mouse strain can be found on [The Jackson Laboratory](#) website and in particular in their Handbook of Genetically Standardized Mice. Chapter 4: Characteristics of Popular Strains of JAX® Mice, Including Reproductive Performance will inform the researcher of which characteristics and welfare risks to expect in their mice.

16

EVENTS TO REPORT TO THE ANIMAL ETHICS COMMITTEE

The Animal Ethics Committee members are aware of the expected events that occur in breeding colonies of laboratory animals (such as cannibalisation of pups, occurrence of a vaginal septum or a prolapsed penis, and the occurrence of hydrocephalus in offspring). The aim of the Adverse Event Report Form is to advise the Animal Ethics Committee of welfare risks that they do not already know about.

An Adverse Event Report submitted for a breeding colony Animal Ethics Approval Certificate could include:

- Unexpected phenotypes associated with a genotype
- A sudden increase in the number of 'natural' deaths that occur in the colony
- An accident, such as a leaking water bottle, that killed pups

Other incidents that should be reported could include a higher than usual cannibalisation rate in breeding animals caused by vibrations due to building work. In such instances, consult with the Officer in Charge of the animal facility. This type of event would likely affect the breeding colonies of many researchers with animals housed in the same location, and therefore the Officer in Charge of the facility would submit the Adverse Event Report Form.

If you think you will need to submit an Adverse Event Report Form contact the Animal Ethics Unit as soon as possible. They will help you provide the information that the Animal Ethics Committee will need to consider the matter, which might include recommending that a necropsy be performed.

CONTACTS

Animal Ethics Unit Coordinator

Tel: + 61 7 336 52713

E-mail: animal.ethics@research.uq.edu.au

Consultant Veterinary Officer

Tel: +61 7 336 53617

E-mail: animal.ethics@research.uq.edu.au

UQ Biological Resources

Tel: 07 3346 4213

E-mail: uqbr@uq.edu.au

[www.uq.edu.au/research/
integrity-compliance/animal-ethics](http://www.uq.edu.au/research/integrity-compliance/animal-ethics)