



## Animal Ethics Application

Application ID : 0000025625  
Application Title : Mice breeding colony for Victoria University of Wellington  
Date of Submission : 24/11/2017  
Primary Investigator : [REDACTED]  
Other Personnel : [REDACTED]

## Intro

### Details of application

Ethics category code\*

Animal

1. Current Committee

Animal Ethics Committee

2. Clearance Purpose code\*

Research

3. Application ID

0000025625

4. School\*

[REDACTED]

5. Application Title\*

Mice breeding colony for Victoria University of Wellington

6. Estimated duration of project. (If ongoing, the maximum approval period is 3 years.)\*

3 years

7. Does this application require formal approval?\*

Yes

No

### Project Details

8. Please list the Principal Investigator (the PI), Co-investigators and Student/Technical Assistants. The Principal Investigator must be academic staff of grade lecturer or above.

If you are not the principal investigator, please delete your personnel entry below and add your name again, with the correct position title..

Please list all personnel involved in this project. Ensure that all are listed with the correct role.

**Please ensure that only one person is listed as Principal Investigator and ticked as Primary**

To add a person, search for their Victoria ID if known, otherwise *either* their first *or* last name (whichever is the most unusual). Click on the magnifying glass to search for results.

Press the **green tick** at the bottom right corner to save the person record.

Add anybody who is involved in this project as:

- Associate Investigator
- Other Researcher
- PhD Student
- Masters Student
- Research Assistant

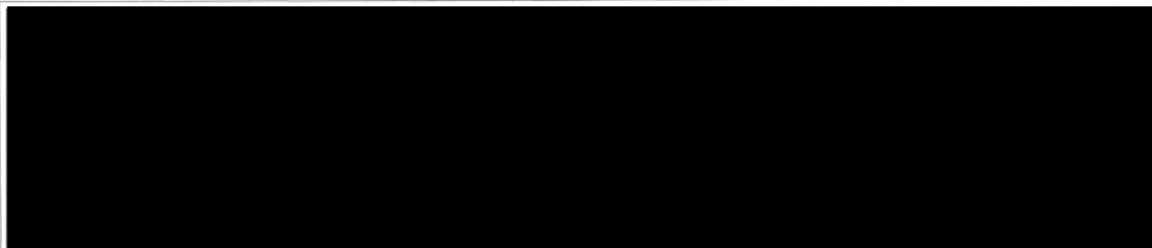
Click on the help button if you are having difficulty adding people to the list.\*

1	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	No
2	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]

	Email Address	[REDACTED]
	Primary?	No
3	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	No
4	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	No
5	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	No
6	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	No
7	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	Yes
8	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	No
9	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]

	Primary?	No
10	Title	[REDACTED]
	Given Name	[REDACTED]
	Surname	[REDACTED]
	Full Name	[REDACTED]
	Position	[REDACTED]
	Email Address	[REDACTED]
	Primary?	No

8a. Please describe the qualifications and experience of each researcher listed above and list any training plans.



Postgraduate students will be appropriately trained by the Principal Investigators or [REDACTED] to care for the mice in their absence (please see attached document - List of additional Research Assistants and Students involved in animal husbandry; the list will be modified if new students get involved).

9. Please list the project funding. \*

Internal

10. Lay summary: (Short paragraph for non-scientists; will be made available to the general public.)\*

We would like to maintain our rodent breeding colony to produce the mice needed for the research experiments approved by the Animal Ethics Committee of VUW.

11. Background, aim and significance of project: \*

Several approved research experiments involving mice are conducted in our facility each year. In order to have enough animals for these experiments, or to have animals showing particular characteristics, we would like to maintain our mice breeding colony.

12. State where the animals will be housed, who will care for them, how they will be maintained, and who will carry out anaesthesia, surgery and euthanasia: (refer [Code Section 5](#))\*

Animals will be housed in the Small Animal Facility (SAF) in the Central Services Building, VUW, until the shift into the new building, Te Toki A Rata (TTR) in 2018. Concerning [REDACTED] group, [REDACTED] primarily and [REDACTED] and/or other appropriately trained postgraduate students, in her absence will care for the animals. They will be maintained on an ad libitum diet and standard 12 hour light-dark cycle. Colony management and euthanasia will be performed by [REDACTED] and [REDACTED] or other trained postgraduates. Due to [REDACTED] being out of the Wellington region during the 2017-2018 Christmas shutdown period, trained personnel (see [REDACTED] group in the list of additional personnel) will be cleaning and monitoring the animals. Following the SAF move to TTR, [REDACTED] is likely to be responsible for breeding activities. Concerning [REDACTED] group, the main carer of the animals will be [REDACTED] lab. All animals will be housed in plastic cages containing bedding, shredded paper for nesting, and a toilet roll for enrichment. They will have free access to food and water, which will be checked 3 times a week and replaced where necessary. The cages will be cleaned and replaced once a week. The euthanasia of these animals will be carried out by all personnel listed in [REDACTED] group (see list of additional personnel). Once in TTR, animals will be housed in individually ventilated cages. The rooms where the animals will be housed will be temperature controlled and kept at 21 degrees Celsius and 55% humidity and standard 12 hour light-dark cycle. Food and water will be freely available. [REDACTED] will perform the breeding activities (selection, pairing, weaning and culling) and care for the mice. The mice cages have a floor area of 484 cm<sup>2</sup> allowing for up to 5 mice of 25 g to be housed together. An exhaust system allows each cage to be ventilated in order to keep low levels of ammonia and CO<sub>2</sub> in the cages, and to reduce the spread of infections and contaminants. By default, all animals will receive enrichment material (chew blocks, cardboard rolls, nesting material).

13. Proposed avenue of publication of research results: (Results from research projects are expected to be published. For on-going projects, evidence of publication is required before approval can be given to renew the application for a further 3 years.)\*

No research result will be obtained from the breeding per se, but experiments for which the breeding colony is necessary will yield results that will be published in international journals.

14. Is this a continuation of an ongoing project?\*

- Yes  
 No

15. Do you require an IDAO form? \*

- Yes  
 No

16. Do you intend to use any of the Standard Operating Procedures for the following? A new page will appear when each option is chosen and the page is saved.

- Biological Sciences
- Malaghan
- Psychology

### Standard Operating Procedures - Malaghan

17. Which of the following SOPs are you using?

SOP 10M - Ethical end point in sick mice

Mouse Grimace Scale Orbital tightening: the closing of the eyelid Nose bulge: a bulging on the bridge of the nose Cheek bulge: a bulging of the cheeks Ear position: ears move back from facing forward to lay on body. Ears rotate outwards and space between the ears increase. Whisker change: whiskers either being pulled back against the cheek or pulled forward to 'stand on end'. Whiskers may clump together. Other signs of illness: - The mouse is hunched over - Eyes are sunken - Ruffled fur - Slow/immobile - Weight loss Behavioral patterns: - Mouse does not socialize with other cage mates - Does not groom/clean itself - Does not interact with any enrichment they are given

### Species justification, DOC and Justification of Use

18. Give the species, strain, sex, age, and source of animals, and state the total number of each species needed for the project. (refer (refer [Code vii](#)))\*

Species: Mice  
 Strains: B6.SJL ptprca, BALB/c, BALB/cByJ-Lpin1fld/J, BALB/cNctr-Npc1m1N/J, DAZL KO mice, FancD2 KO mice, 12HI-RAstat-GFP, 4HI-GFP-ZHI-TOM-RAstat, 2D2, IL-4Ra-/-, C57BL/6J  
 Sex: males and females  
 Number: up to 2600 animals

19. Does your project require DOC approval?\*

- Yes
- No

20. Justification of animal use. Explain why the proposed use of animals is unavoidable, what alternative approaches are available, and how the number of animals used will be minimised. (State prior history of animals, and provide statistical or biological justification of numbers.) (refer (refer [Animal Welfare Act 1999 s 100](#)))\*

The only purpose of our breeding colony is to produce the mice needed for the experiments conducted in our facility and approved by the Animal Ethics Committee.  
 Because we need animals showing standard responses to experimental manipulations and treatments, we need them to be bred in standard conditions in a colony. The breeding colony needs to be on site in order to have enough animals for experiments, and to have animals showing particular characteristics, such as the KO mice mentioned above. Breeding mice in situ also means they don't need to go through long hours of transport.

### Animal Manipulation

21. Pain classification of project: (Please indicate the grade(s) of your manipulation(s):\*

- Grade A - no impact
- Grade B - little impact
- Grade C - moderate impact
- Grade D - high impact
- Grade E - very high impact

22. Experimental design of project: (give overall design, including details of protocols involving animals, including details of experimental and control groups of animals, and state if the method to be used is standard practice of a new approach. If applicable, give details of risk management and containment procedures.) If the details exceed a character limit of 4000, please upload details as a document to the Documents page. \*

lab: Once at 8 weeks of age or above, C57BL/6J mice will be placed into a breeding cage following one of 2 mating combinations based on need and numbers of breeding age mice. We will utilise a breeding pair combination in which one male and one female are placed in a cage together, and remain together for the duration of their breeding time to maximise the offspring produced. We will also utilise mating trios, in this situation one male will be placed in a breeding cage with 2 females to increase the production of offspring. Where possible the trio will be left in the cage to allow for the mothers to co-parent the pups. If necessary the mothers will be separated after one week with the male to give birth and raise the pups separately, once the pups are weaned at 3 weeks old the females will be placed back with the same male to produce their next litter.  
 All DAZL and FancD2 mice will be ear-notched at 2-3 weeks of age for the purpose of identification and genotyping.

22a. What, if any, statistical advice have you sought for this application?\*

NA

23. Surgical procedures: (give details for each type of operation) (Refer [Code, Section 5](#))\*

NA

24. Provisions for post-manipulation recovery and care - definitions of endpoints - whether based on the aims of the study, humane considerations, or death of animal : (refer [Code, Section 5](#))\*

Breeding mice will be euthanised at 7-8 months old, if they produce consistently small litters (2-3 pups), or if they do not produce a litter within a 2 month time period. Mating pairs/trios' will also be monitored for signs of aggression and/or injury towards the female(s), aggressive mice will be immediately removed from the breeding programme. Animals losing 15% of their weight overnight for no explainable reason (for ex. empty water bottle) or showing signs of illness will be euthanised. DAZL and FancD2 mice: Ear-notching has a low pain impact (as judged by animal behaviour provided it is performed on unweaned mice). The KO animals of both strains >2 months of age begin to develop cancerous lesions. Animals between 5 months and 1 year of age will develop high grade serous ovarian carcinomas and therefore need to be monitored for tumour growth by visual inspection and other signs of pain as per SOP 10M. Checks for changes in behaviour, coat appearance and abdomen distension are made on a regular basis (fortnightly). To date, <0.1% of KO mice has been euthanised due to cancer progression.

25. What is the fate of animals at conclusion of study: (refer [Code, Section 5](#))\*

Euthanasia

26. Does your research involve anaesthesia or euthanasia?\*

- Yes  
 No

26a. Anaesthesia or euthanasia procedures: (give specific drugs, doses, and routes of administration) (refer [Code 5 xi, xii](#))

Animals will be sedated with carbogen (CO2/O2 mix), then euthanised with CO2 / cervical dislocation.

**Documents**

27. Upload documents to this page.

Description	Reference	Soft copy	Hard copy
Research Design	List of additional Research Assistants and Students involved in animal husbandry - Mice.docx	✓	

**Declaration**

28. I am aware that this project is undertaken within the provisions of the Animal Welfare Act 1999, I have read the Victoria University of Wellington Code of Ethical Conduct for the Use of Live Animals for Teaching and Research, and I agree to abide by all the conditions contained in these two documents. In the event of this application being approved, I will promptly inform the Animal Ethics Committee of any subsequent unforeseen change or planned modifications to the project, giving explanations for all such changes.

I agree to maintain accurate records of all animals used (refer [Code, Section 4 \(c\)](#)) and to make these records available promptly to the Executive Officer of the Animal Ethics Committee. If this is a modification, I agree that the proposed changes will be carried out in conformance with the details approved.\*

- Yes  
 No

29. Please add the Head of School (or delegate) who should be notified about this application. This will be your own Head of School, or the person in your School responsible for Ethics applications. **Please check with your School administration team if you are unclear who should be assigned this role.**

**Once you've searched for your Head of School/delegate, click on the green tick to add them, and then also save the application before submitting.**

\*

1	Given Name	██████████
	Surname	██████████
	Full Name	████████████████████
	Position	██

**AEC Administration (committee use only)**

AEC Internal File Reference

*This question is not answered.*

Date Approved

14/12/2017

Date IDAO Approved

14/03/2019

AEC notes on application

18/1/19: Update of mouse strains in facility  
Feb 2019: Mouse use for 2018 reported to MPI.  
7/3/19: Modification 2 approved  
31/5/19: Modification 3 approved (minor amendment)  
3/7/20: Modification 4 (additional mouse strains)

This project is approved until this date of expiry

01/01/2021

Date Reapproved

*This question is not answered.*

### Modifications and Extensions

1. Which of the following are you applying for? Click all that apply.

- Modification  
 Extension  
 Personnel change or addition

1a. Describe the proposed changes in detail. State the section(s) of the existing approval affected, and the proposed differences, including information on animal numbers and whether the modification involves replacement of already approved animals or addition of extra animals.

Addition of recently imported or transferred mouse strains into the TTR animal facility:

B-arrestin k K/O  
Oprm-1  
PDGFr-alpha GFP  
B6.SJ.OT1

1b. State the number of additional animals needed for the modification, if any

No change

1c. Outline why the changes are needed, indicating the main benefits of the proposed changes.

The update keeps the list of animals held in the facility current

1d. Provide information on the likely animal welfare implications of the proposed changes

No change

1e. Is there a change in the impact/pain classification?

No change

1f. If additional animals are requested or reassigned in this modification, please provide details

No change

1h. Indicate how urgently this application needs to be processed

N/A

1i. How long is the extension you need?

N/A

Subsequent Amendments (further requests after initial amendment request has been approved) If you have already had an extension or amendment in the past, please answer the questions below.

Do you have a second modification or extension to make?

- Yes

### Second Modification and/or Extension

2a. Which of the following are you applying for?

- Second Modification  
 Second Extension  
 Second Change or Addition of Personnel

2b. Describe the proposed changes in detail. State the section(s) of the existing approval affected, and the proposed differences, including information on animal numbers and whether the modification involves replacement of already approved animals or addition of extra animals.

We would like to add SOP 42M – Non-survival caesarean section to the existing protocol.

Caesarean rederivation (also referred to as hysterectomy derivation) is a commonly used procedure to obtain specific pathogen free (SPF) mice from an infected strain, or from mice with an unknown health status. As there is a projected increase in the importation of genetically modified animals/strains of animals from international collaborators and facilities, it is important that these animals adhere to our (Victoria University of Wellington) facility standards. Rederivation via caesarean of these animals is considered the safest and most effective procedure to protect our animals from possible infection and prevent misinterpretation of experimental results.

SPF foster females will be mated, and the donor females will then be mated 2-3 days later. The caesarean section will be performed on the donor one day before expected parturition. The donor mother will receive 0.25mg/kg Celestone Chronodose (betamethasone) via subcutaneous administration the day before and on the day of the caesarean section. She will be euthanized by cervical dislocation, dipped in dilute iodine solution and the uterus will then be removed. The entire uterus will be placed in a warmed pot of dilute iodine solution for one minute. The amniotic membrane will be torn using cotton swabs, and the nostrils and mouth of the pups cleared. The pups will be placed on a heat pad, be gently dried and stimulated until they are breathing well and gain a healthy pink colour indicating good oxygenation. The pups will then be transferred to the foster mother with a known health status as soon as possible.

2c. State the number of additional animals needed for the modification, if any

We will not be increasing animal numbers from the original application.

2d. Outline why the changes are needed, indicating the main benefits of the proposed changes.

Currently, we have obtained a line of PdgfR $\alpha$ -H2B-eGFP mice from Monash University, Melbourne. These mice have come from a facility where the Proteus species is present in all areas of their animal housing facility. This pathogen is not tolerated in our facility and therefore these mice cannot be accepted. At this time, we are unable to source this genetic line from an institution with higher health standards. As such, this procedure is less invasive to the animal in comparison to other assisted reproductive techniques.

2e. Provide information on the likely animal welfare implications of the proposed changes

There is a risk that cross-contamination occurs during the procedure which will mean that the pups will need to be culled, as they are not to the standards of our facility. To mitigate this risk, surgical tools will be sterilised, aseptic technique will be followed including using separate tools for accessing the uterus and dissection of the uterus.

There is also a risk that the foster mother will reject the pups. We will rub the scent of the foster mother's cage on the derived pups before introducing them to the cage. We will also closely monitor the pups and check for signs of feeding (i.e. milk spot and increase in size)

2f. Is there a change in the impact/pain classification?

No change

2g. If additional animals are requested or reassigned in this modification, please provide details

N/A

2h. Indicate how urgently this application needs to be processed

March AEC meeting

2i. Please list the new personnel here

[Redacted]

2j. Please describe the qualifications and experience (or any training plans) of the new personnel.

[Redacted]

Do you have a third modification or extension to make?

Yes

### Third Modification and/or Extension

3a. Which of the following are you applying for? Click all that apply.

- Third Modification
- Third Extension
- Third Change or Addition of Personnel

3b. Describe the proposed changes in detail. State the section(s) of the existing approval affected, and the proposed differences, including information on animal numbers and whether the modification involves replacement of already approved animals or addition of extra animals.

Addition of a new mouse strain (Ts65Dn) plus controls (B6EiC3SnF1/J) to the list of strains held by facility  
Anticipated breeding of males and females to produce up to 500 offspring per year.

3c. State the number of additional animals needed for the modification, if any

Up to 500 animals per year



- 3d. Outline why the changes are needed, indicating the main benefits of the proposed changes.
- New grant exploring aneuploidy requires the purchase of a new mouse strain (Ts65Dn (B6EiC3Sn-Rb(12.Ts171665Dn)2Cje/CjeDnJ) mice) and equivalent controls (B6EiC3SnF1/J) from the [REDACTED] laboratory for the purpose of generating Trisomy 21 embryos.
- 3e. Provide information on the likely animal welfare implications of the proposed changes
- These mice will euthanised in a barren state for collection of oocytes or one day after fertilisation for collection of zygotes. Mice will be euthanised using standard procedures (under tissue collection protocol 25766).
- 3f. Is there a change in the impact/pain classification?
- No. These mice are a model of Downs Syndrome and we do not anticipate a change in the impact/pain classification/
- 3g. If additional animals are requested or reassigned in this modification, please provide details
- Purchase of a new mouse strain (Ts65Dn (B6EiC3Sn-Rb(12.Ts171665Dn)2Cje/CjeDnJ) mice) and equivalent controls (B6EiC3SnF1/J) from [REDACTED] lab. Up to 500 animals per year.
- 3h. Indicate how urgently this application needs to be processed
- Very urgent
- 3i. Please list the new personnel here and describe their qualifications and experience (or any training plans).
- [REDACTED]
- 3j. Please describe the qualifications and experience (or any training plans) of the new personnel.
- [REDACTED]
- Do you have fourth modification or extension to make?
- Yes

#### Fourth Modification and/or Extension

- 4a. Which of the following are you applying for? Click all that apply.
- Fourth Modification  
 Fourth Extension  
 Fourth Change or Addition of Personnel
- 4b. Describe the proposed changes in detail. State the section(s) of the existing approval affected, and the proposed differences, including information on animal numbers and whether the modification involves replacement of already approved animals or addition of extra animals.
- Addition of new mouse strains to list held by facility:  
 JAX 001924 - B6EiC3Sn a/A-Ts(17<sup>16</sup>)65Dn/J  
 JAX 001875 - B6EiC3SnF1/J  
 JAX 016959 - B6.129(Cg)-Foxp3<sup>tm4</sup>(YFP/icre)Ayr/J  
 JAX 030076 - B6;129-Oprk1<sup>tm2.1Kff</sup>/J
- 4c. State the number of additional animals needed for the modification, if any
- No change
- 4d. Outline why the changes are needed, indicating the main benefits of the proposed changes.
- Recently imported for use in protocols approved by the AEC
- 4e. Provide information on the likely animal welfare implications of the proposed changes
- No changes
- 4f. Is there a change in the impact/pain classification?
- No changes
- 4g. If additional animals are requested or reassigned in this modification, please provide details
- No changes
- 4h. Indicate how urgently this application needs to be processed
- N/A

Mice breeding colony for Victoria University of Wellington – List of additional Research Assistants and Students involved in animal husbandry

- [REDACTED] group:

Name	Position	Email address
[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]

[REDACTED]

- [REDACTED] group:

Name	Position	Email address
[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]
[REDACTED]	[REDACTED]	[REDACTED]

[REDACTED]