

Mouse Genotyping Service Sample Submission Guidelines

Online Sample Submission Portal

Please use our Online Sample Submission Portal on our website at:

<https://gmg-submit.gimr.garvan.org.au/#/login>

In-depth instructions are available here:

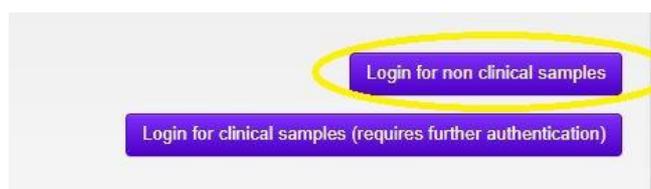
https://www.garvan.org.au/research/capabilities/molecular-genetics/documents/gm_15_v1.pdf

Video instructions are available here: <https://www.youtube.com/watch?v=8eL-JJ8SIGU>

Further information to our services can be found here: www.garvan.org.au/gmg

Quick Guide: Using the Sample Submission Portal

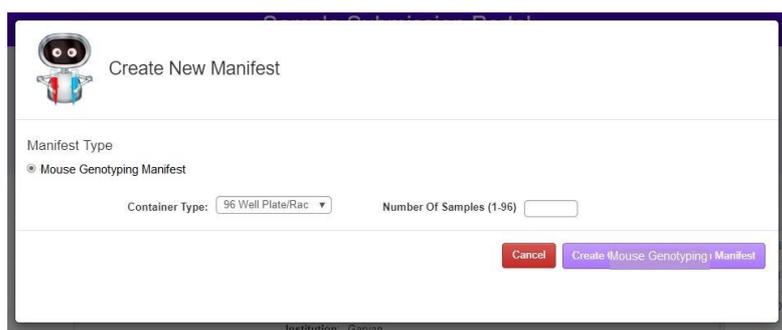
New clients need to create an account (please follow instructions after clicking on the “Create Account” button on the landing page), existing clients log into their account by selecting the button “Login for non clinical samples”:



After logging in you the next screen will show your account. Here you can submit new samples and view previous submissions. To submit a new manifest for mouse genotyping, select “New Mouse Genotyping Manifest” from these options:



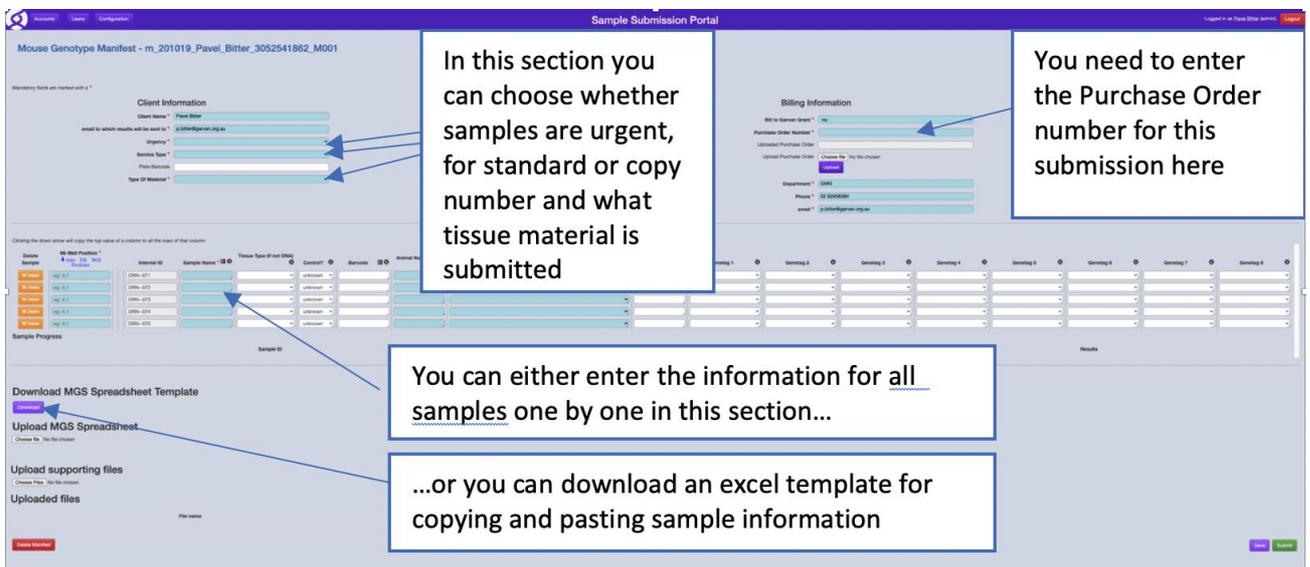
In the following popup window, select the container type and the amount of samples that will be submitted. We strongly prefer that samples are submitted in barcoded tubes in 96-well racks. We can supply these (racks, lids, tubes, barcodes) to you at no additional cost if you email us at mgs@garvan.org.au to request them.



After clicking “Create Mouse Genotyping Manifest”, the new Manifest will open (see below). Please fill out the client and billing information fully and include a Purchase Order (PO) if your institution requires one to process invoices. If you can confirm that your institution does not need a PO in order for invoices to be paid, you can fill this field with a “0”. The PO can also be updated after the manifest has been submitted (but must be before results are released).

There are two ways to fill out the sample form; either on the webpage (this may be more convenient for you if you are only submitting a few samples), or via the excel sheet form under “Download MGS Spreadsheet Template”.

When filling out the webpage form, once a mouse line is selected, the genetag dropdown menu will show only genetags that are associated with that line. Please let us know if any of your lines are missing, or if there are any genetags that you would like added to a line.



Client Information
In this section you can choose whether samples are urgent, for standard or copy number and what tissue material is submitted

Billing Information
You need to enter the Purchase Order number for this submission here

Sample Submission
You can either enter the information for all samples one by one in this section...
...or you can download an excel template for copying and pasting sample information

The downloaded template can be filled out as follows (see below). Please provide unique sample names (you can use the sample barcode as the sample name as well, to ensure uniqueness).

Well Plate Position / Tube Number	Sample Name	Tissue Type	Control	Barcode	Animal Number	Mouse Line	Comment
1A	1	Tail			123	1D1A	
1B	2	Tail			124	1D1A	
1C	3	Tail			125	1D1A	
1D	4	Tail			126	1D1A	
1E	5	Tail			127	1D1A	

In the downloaded template sample names can be entered by typing or via copying and pasting

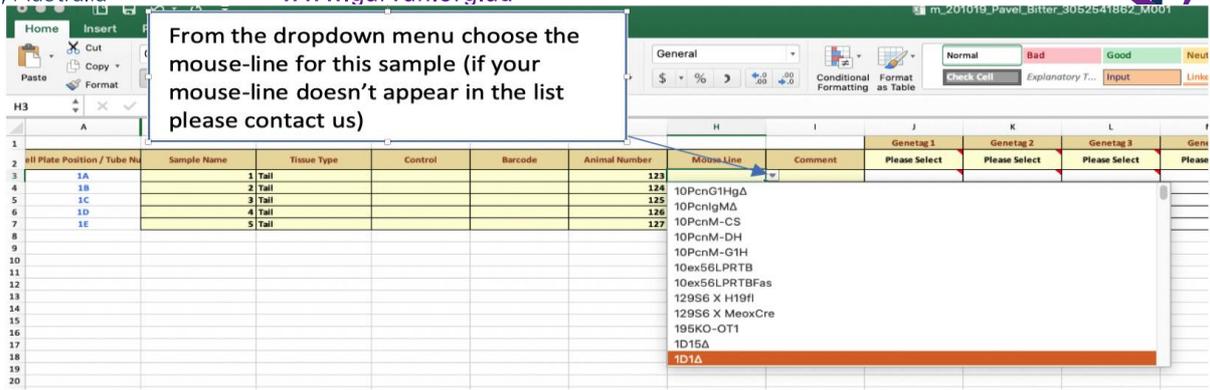
The tissue type can be selected here

If the sample is a control sample please indicate this here

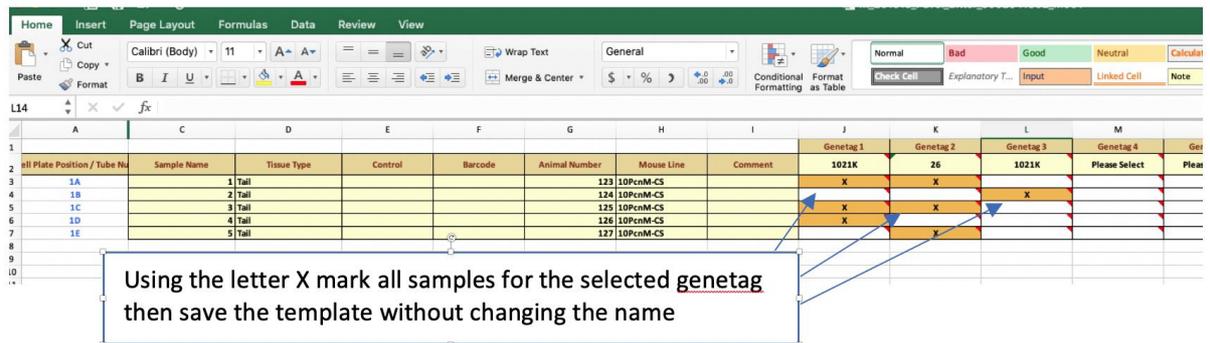
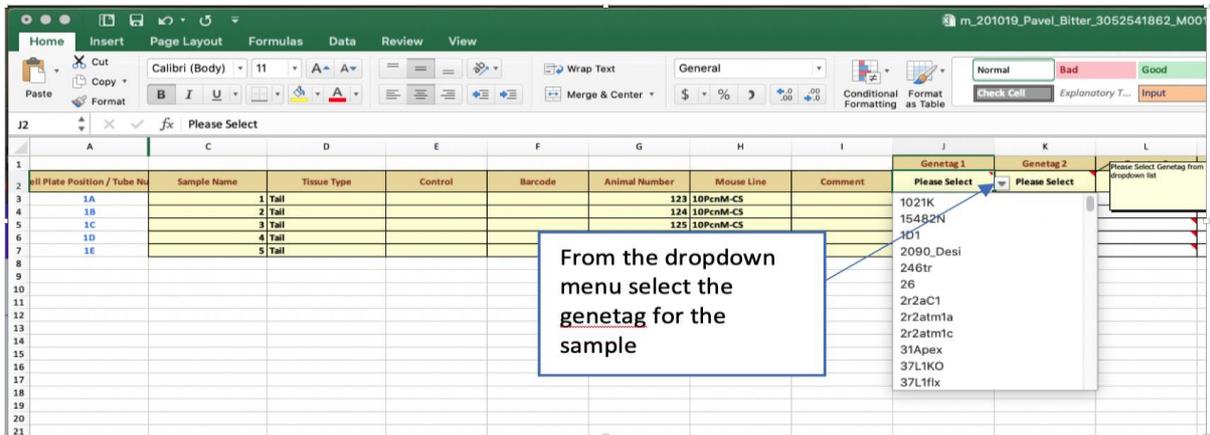
If the samples are labelled with barcodes please enter the barcodes here

The animal numbers can be entered in these fields

Similarly to the webpage form, you will be able to select your mouse lines from the dropdown menu.

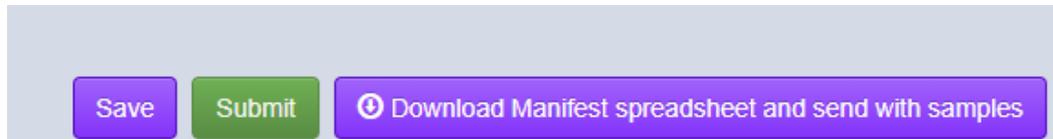


After your mouse line has been selected, please select what genetags you would like run for your samples. This is achieved by selecting the relevant genetag from the dropdown menu and then typing "x" in the row for what samples you want run with it (see the images below). You will see every genetag in this view, but please ensure you only select genetags that are associated with the line you have selected.



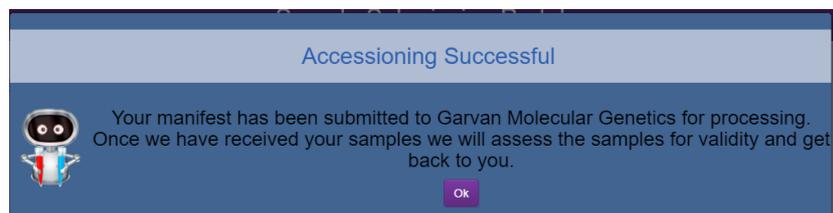
Once you have completed this form, save it (do **not** change the file name) and upload it by selecting "Choose File" under the "Upload MGS Spreadsheet" header. If there are any issues with the excel sheet, the system will tell you in an error message. If anything is unclear in this message or you are unable to rectify it, please let us know. Once the sheet is successfully filled out and selected, click the "Upload" button that appears and select "ok" once the upload has completed. After this, you will see the webpage form will be updated with the information you have provided in the spreadsheet.

When you are ready, scroll to the bottom of the page, and select the green “Submit” button.



If there are any issues with your submission, the manifest will not be submitted. Any fields with missing information or that have been filled out incorrectly will be highlighted with a red outline. Hovering over this will supply further context on what needs to be fixed.

Once the form has been submitted successfully, you will see the following popup:



Standard Genotyping

Our standard mouse genotyping is based on high resolution meltcurve analysis. We require **controls with known genotypes** (tissue or DNA) to perform this analysis. Exceptionally we can deduct the genotype without controls if you take responsibility for the results. To start a new genotyping assay please fill in the **New Project Submission Form** which you can download from our webpage www.garvan.org.au/gmq > [mouse genotyping](#).

Submitting tissue samples

If you submit ear clips samples please make sure they are **at least 3mm² large** (you can submit more than one earclip of the same mouse in one tube). Please note, if the tissue size is too small we cannot extract sufficient DNA and will have to reject that sample. If you submit tail tips please submit them in a range of 3 to 5mm. Please make sure samples are not cross contaminated by other tissue and that the tissue in the well corresponds with the sample submission manifest (no sample confusion).

Copy number analysis via real-time PCR

If heterozygous and homozygous transgenic mice cannot be differentiated via standard PCR we can run a real-time PCR and separate the heterozygous from the homozygous samples via real-time Ct values, see Shitara H. “Simple method of zygosity identification in transgenic mice by real-time quantitative PCR”, *Transgenic Res.* 2004 Apr;13(2):191-194.

Quality of submitted DNA samples

For this very sensitive real-time PCR we will amplify the sample DNA 5 times with a target and a house keeper gene, the average Ct of the target gene is normalized against the house keeper and compared to the submitted controls, one cycle difference in the Ct indicates the difference from het to hom. For this analysis we can only accept column cleaned DNA.

Sample volume and concentration

We will need at least **100ul of DNA** with a **concentration of >50ng/ul**. The concentration of the submitted samples must be adjusted to be the same concentration for all samples (+/- 5%) including the controls. We can prepare your samples for you for an added service fee. Please indicate in the sample submission form if you want us to perform the equilibration. The 260/280 ratio must be between 1.7 to 2.3 and the 260/230 ratio must be between 1.9 to 2.4. These are our acceptance

384 Victoria Street
Darlinghurst NSW 2010
Sydney, Australia

T +61 2 9295 8100
F +61 2 9295 8101
www.garvan.org.au



criteria for DNA that will be accepted to the service. DNA that fails these criteria can be sent to us and we can perform genotyping but we will not take responsibilities of the results.

Controls

We will need at least one control which is either Het or Hom, ideally both, WT samples are of no use. The DNA of these control samples should be extracted at the same time as the samples that are submitted.

Turnaround time

The turnaround time for this analysis can take up to two weeks although usually and especially when requested results are available earlier.

Sending of Samples

We prefer sending you our tubes in racks with barcode labels. You can then use these tubes to send us your tissue samples.



Alternatively submit your samples in 1.5ml Eppendorf tubes (3810X PCR clean 1.5ml, Catalogue # 0030125.215 Eppendorf). Please position these samples in the rack that we send you according to your online sample submission manifest.

We recommend to cool DNA or tissue samples when sending samples with mail or courier (cooling packs, ice or dry ice). However if the transport can be arranged overnight it is acceptable to send samples without cooling in express post envelopes or via courier.

Shipping Address:

Our shipping address is:

Garvan Institute

Loading Dock
West Street (off Burton Street)
Darlinghurst, NSW, 2010

Dock times: 8am to 4pm

Phone: 02 9295 8640

Pricing

Pricing information is available on our website at www.garvan.org.au/gmg > Molecular Genetics Shop.

Results:

Our online sample submission portal will send you an email with a link to download your results within within an approximate turnaround time of 48-72h for standard genotyping (copy number results take several working days). If you are a Stuart software customer, results will also be uploaded into Stuart for you.

Contact us:

Garvan Molecular Genetics

internet: www.garvan.org.au/gmg

email: gmg@garvan.org.au

Pavel Bitter (Molecular Genetics Facility Manager)

email: p.bitter@garvan.org.au

phone: 02 92958384

Email: gmg@garvan.org.au

<http://www.garvan.org.au/research/capabilities/molecular-genetics>

