Enabling Precision Medicine

Phenomics Australia
Enabling precision medicine

NCRIS
National Research Infrastructure for Australia
An Australian Government Initiative

Small and large scale drug screening

Enabling discovery and driving translational medicine with high throughput technologies

VCFG Victorian Centre For Functional Genomics

Peter Mac Peter MacCallum Cancer Centre Victoria Australia

Small and large scale drug screening at VCFG

Why do a High Throughput screen?

- Why not? Why spend months doing everything by hand, adding human error with every plunge of your pipette. By using our state of the art equipment you accurately and reproducibly finish your screen in a fraction of the time in 96 or 384 well formats.
- Ask big picture questions, or small scale discovery, use plate reader, FACs or imaging end points.

What can I screen?

- It's very flexible. Anything from small numbers of BYO drugs, to drug synergy interactions, to a boutique library of your choice or much larger collections.
- The VCFG has access to many libraries through Compounds Australia at Griffith University; including the FDA (>5000), Kinase Inhibitors, Metabolic regulators, Epigenetics, Scaffold collection, Baell-MIPS defined collection, CSIRO and NatureBank.
- Opportunities for pilot screening data for grants, or as a precursor to apply to the NDDC.

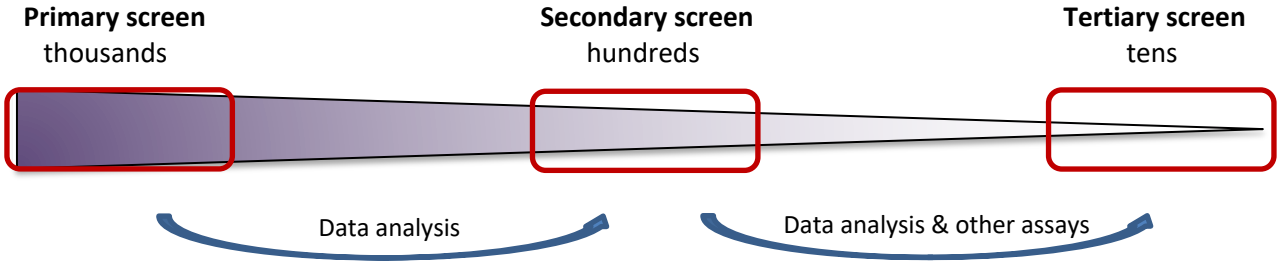
What types of screen can I do?

The readout for a screen can be as simple as a plate reader-based viability or reporter readout, or imaging-based DAPI stained viability and cell cycle determination, through to highly multiplexed cell morphology and phenotypic profiling. All this can be done in both 2D and 3D formats.

What are the benefits of a screen?

High Throughput screening provides unbiased discovery from a full library of compounds through to validated hits. You can triage your hits by clustering using imaging to find novel connections to unrelated compounds.

Concept of a screen



How do I prepare for a screen?

Before you jump in to a screen you will need to optimise the following:

- Cell density; ~80% confluence at your end point
- Test positive and negative controls
- Work out a time line for dosing and number of doses required
- Plan ahead for a 3 week lead time with Compounds Australia
- Test growth kinetics on the IncuCyte
- Quality control metrics, %CV and Z'factor

What equipment will I need to be trained on for my screen?

You will need training on:

- BioTek EL406 for cell dispensing/media changing/fixing/staining.
- Tecan D300E for small scale drug dosing and delivery of controls.
- Cytation 5/10 or Cellomics CX7 microscopes for end point analysis (plater reader or imaging).
- Incucyte SX5 for cell density optimisation.
- We will run the large robots for plate-scale liquid transfers.

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What is the workflow for a screen?

A generalised screen might look like this

Day 0	Seed cells (using the BioTek EL406 dispenser for 2D or Janus robot for 3D)
Day 1	Dose cells with compounds (Janus and/or the Tecan)
Day 2	Incubate with compounds (grow in LiCONICs incubator)
Day 3/4	Endpoint assay - fix/stain/image or plate reader based assays

Longer assay options, additional dose time points, regular Bright field imaging is also possible

What controls do I need for a screen?

	1	2	23	24	
A	Media	DMSO	Cisplatin 0.1	DMSO	A
B	Paclitaxel 10	Cisplatin 1.0	DMSO	Media	B
C	DMSO	DMSO	Doxorubicin 1.0	Paclitaxel 0.1	C
D	Doxorubicin 0.1	Doxorubicin 10	DMSO	DMSO	D
E	DMSO	Paclitaxel 0.1	DMSO	DMSO	E
F	DMSO	Cisplatin 10	Media	Cisplatin 1.0	F
G	Paclitaxel 1.0	Paclitaxel 0.1	Paclitaxel 10	DMSO	G
H	Doxorubicin 1.0	DMSO	DMSO	Doxorubicin 0.1	H
I	DMSO	DMSO	Media	DMSO	I
J	Cisplatin 10	Media	DMSO	Paclitaxel 1.0	J
K	DMSO	DMSO	Doxorubicin 10	DMSO	K
L	Doxorubicin 10	Media	DMSO	DMSO	L
M	Media	DMSO	DMSO	Cisplatin 0.1	M
N	Cisplatin 1.0	Doxorubicin 0.1	Cisplatin 10	Doxorubicin 1.0	N
O	DMSO	Paclitaxel 10	Paclitaxel 1.0	DMSO	O
P	DMSO	Cisplatin 0.1	DMSO	DMSO	P

- Lots of controls are needed on every plate. Controls should be randomized and balanced on the left and right sides
- Controls include:
  - Vehicle
  - Untreated
  - Positive/negative biological controls at varying doses
  - VCFG in house technical control compounds

How long will a screen take?

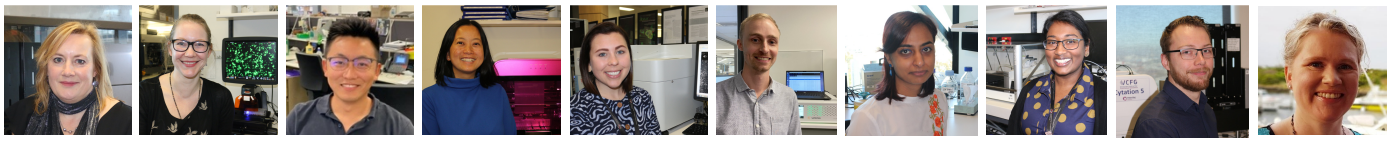
Time is determined by the number of compounds, doses, cell lines, replicates and complexity of your assay. The maximum number of cells plates (depending on endpoint assay) is 30 assay plates per run.

How much will it cost?

There are lots of elements to a screen, we'll work out the best approach for your project and then generate a ball park figure.

The VCFG team – a collaborative and innovative partnership

highly experienced, driving innovative technology and method development, housing large reagent and resource collections, enabling complete end to end service, researcher focused, open access to everyone.



A/Prof Kaylene Simpson - Head, project management, grant support
Dr Susanne Ramm - 2iC, 3D organoid characterization, screening, analysis
Dr Mark Li - 3D screen support and fee for service, analysis
Jennii Luu - Lab manager, automation specialist, 2D and 3D platforms
Karla Cowley - High content imaging and analysis, 2D platform
Dr Henry Beetham - MRFF stem cell project, 2D screens and analysis, CRISPR
Dr Twishi Gulati - CRISPR screening, iLAB management, Business Development
Arthi Macpherson - Equipment training, new instrumentation, PRIME management
Robert Vary - Equipment training, screen support, fee for service projects
Louise Scerri - Administrative support

You can find us on Level 11, Cluster 6, VCCC building - Come and chat any time!

https://www.petermac.org/research/core-facilities-and-services/victorian-centre-functional-genomics

https://research.unimelb.edu.au/facilities-and-resources/research-infrastructure/victorian-centre-for-functional-genomics

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