

High Throughput Screening Facility: Update and Upgrades 2016

SUPERLAB SERIES

PETER BANKS

Outline

Introduction

High Throughput Screening

Robotics – **New and old**

Experiments – **Quantitative Fitness Analysis**

Data Analysis

Random Colony Picking

Drug Screening – **MRCT libraries**

Costs

New Robotics – **Reliance**



Introduction

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2011- High Throughput Screening Facility

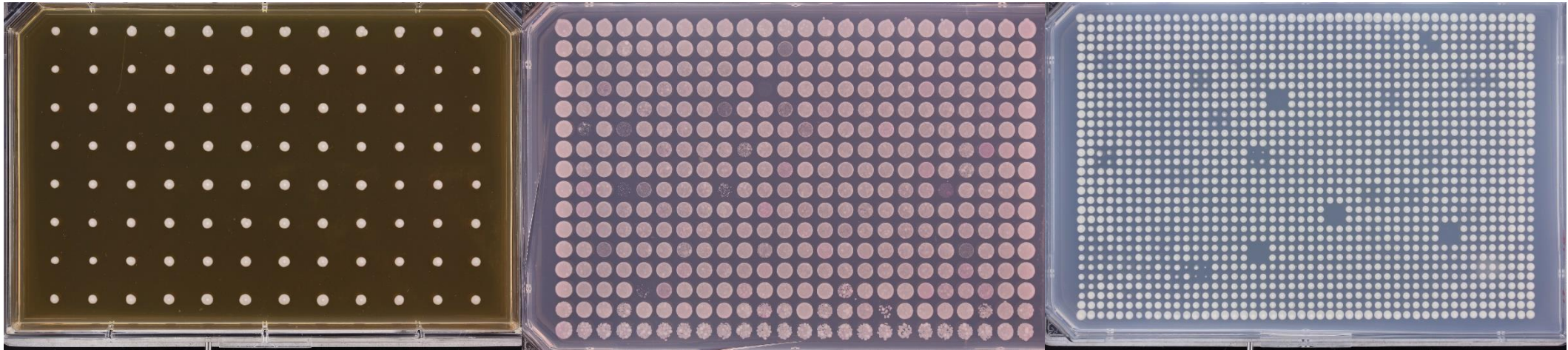
- Equipment
- Reagents
- Expertise

2016 – £400,000 from the Research Infrastructure Fund (RIF)



High Throughput Screening

Transfer manual bench top assays to automated robotic assays commonly in 96, 384 or 1536 format



96 well format

384 well format

1536 well format

S&P BM6 and BM3 colony pinning robots

Housed on 2nd floor Leech and 2nd floor Cookson

Uses metal 96, 384, 1536 and cherry pick pin tools

- low consumable costs

Replicates microbial colonies from agar to agar

- Rearranging and cherry picking libraries

Inoculate microbial colonies from agar into liquid

- Quantitative Fitness Analysis
- Freezing Libraries

Pin microbial colonies from liquid onto solid

- Thawing frozen stocks

High capacity

- 180-240 plates in one run

Slow but robust and easy to use



Singer ROTOR Colony Pinning Robot

Housed on 2nd floor Cookson

Uses plastic 96, 384, 1536 disposable pin tool

- Associated consumable costs

Replicates microbial colonies from agar to agar

Inoculate microbial colonies from agar into liquid

- Quantitative Fitness Analysis
- Freezing Libraries

Pin microbial colonies from liquid onto solid

- Thawing frozen stocks

Low capacity

- 5 plates in one run

Very quick and easy to use



Biomek FXP Liquid Handling Robots

2 systems – 2nd floor Leech and 2nd floor Cookson

1- 96 disposable plastic tips

- Dilution of microbial cultures
- Inoculation of culture from frozen stocks
- Freezing down microbial culture in glycerol – 96 or 384
- Cherry picking

96 well pin tool with free floating pin

- Spotting 96 well plates cultures – Quantitative Fitness Analysis

Very quick and easy to use

High capacity

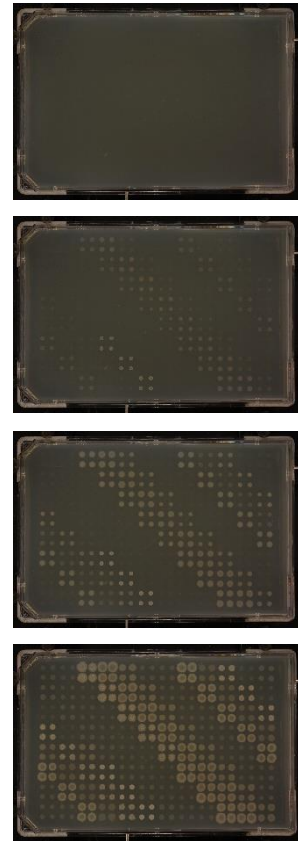
- Deck space for 16 plates
- Cytomat Plate hotel 190 plates in one run



S&P Automated Imaging Systems

Two temperature controlled automated imaging systems

- 20°C-37°C
- Capacity of 160 – 190 plates
- Cycle time of 2 minutes – 1 day
- Run for days
- High quality agar plate images



Ancillary Items

S&P Manual Imager

- High resolution images of round or rectangular agar plates

Singer Serial Filler Plate Pourer

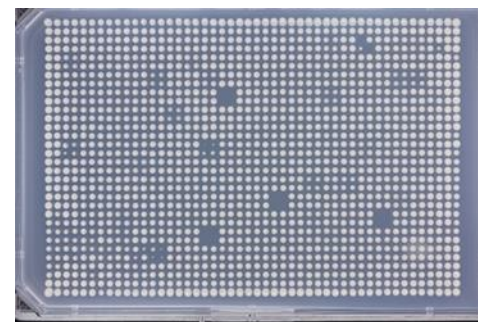
- Round or rectangular plates
- Aseptically dispense up to 2L of media in one run

Matrix Plate Filler

- Dispense 1-2000ul
- 96 or 384 well plates

GreenLab rectangular plates

- Washable and autoclavable plates
- Huge cost saving – Nunc plates are £2+

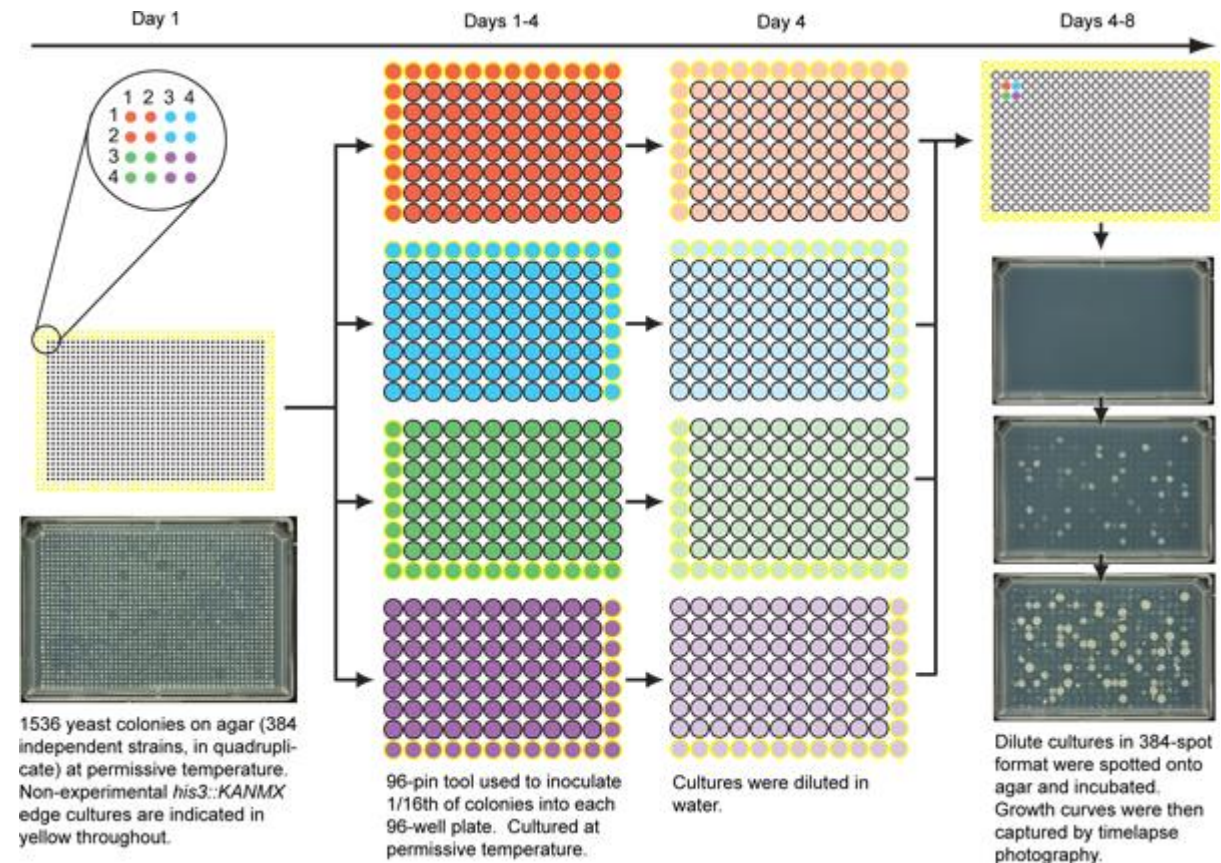


Quantitative Fitness Analysis

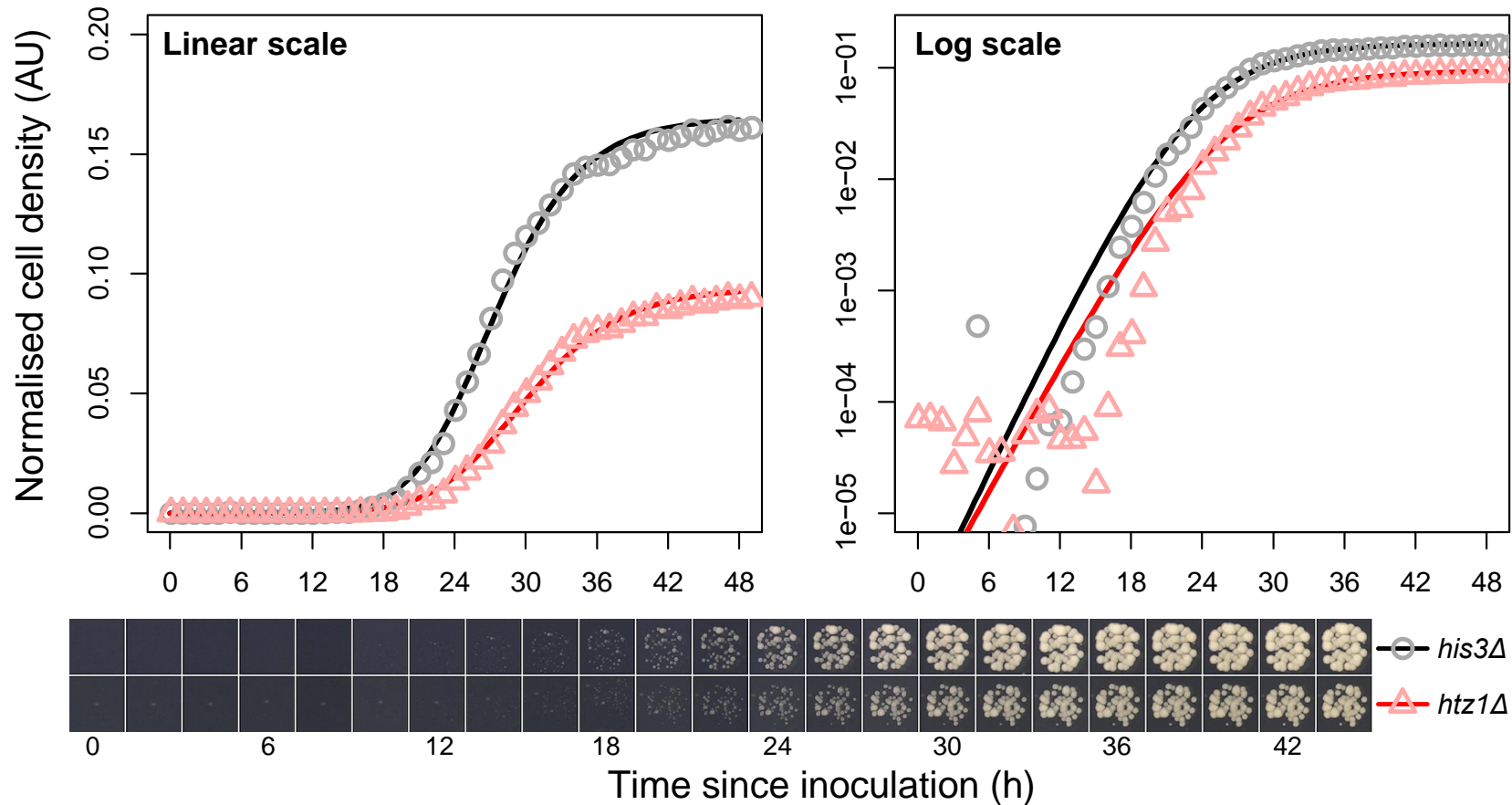
Quantitative Fitness Analysis (QFA) is a complementary series of experimental and computational methods for estimating microbial culture fitnesses

- More phenotypes
- Drug screens
- Temperature sensitive screens

<http://www.jove.com/video/4018/a-quantitative-fitness-analysis-workflow>

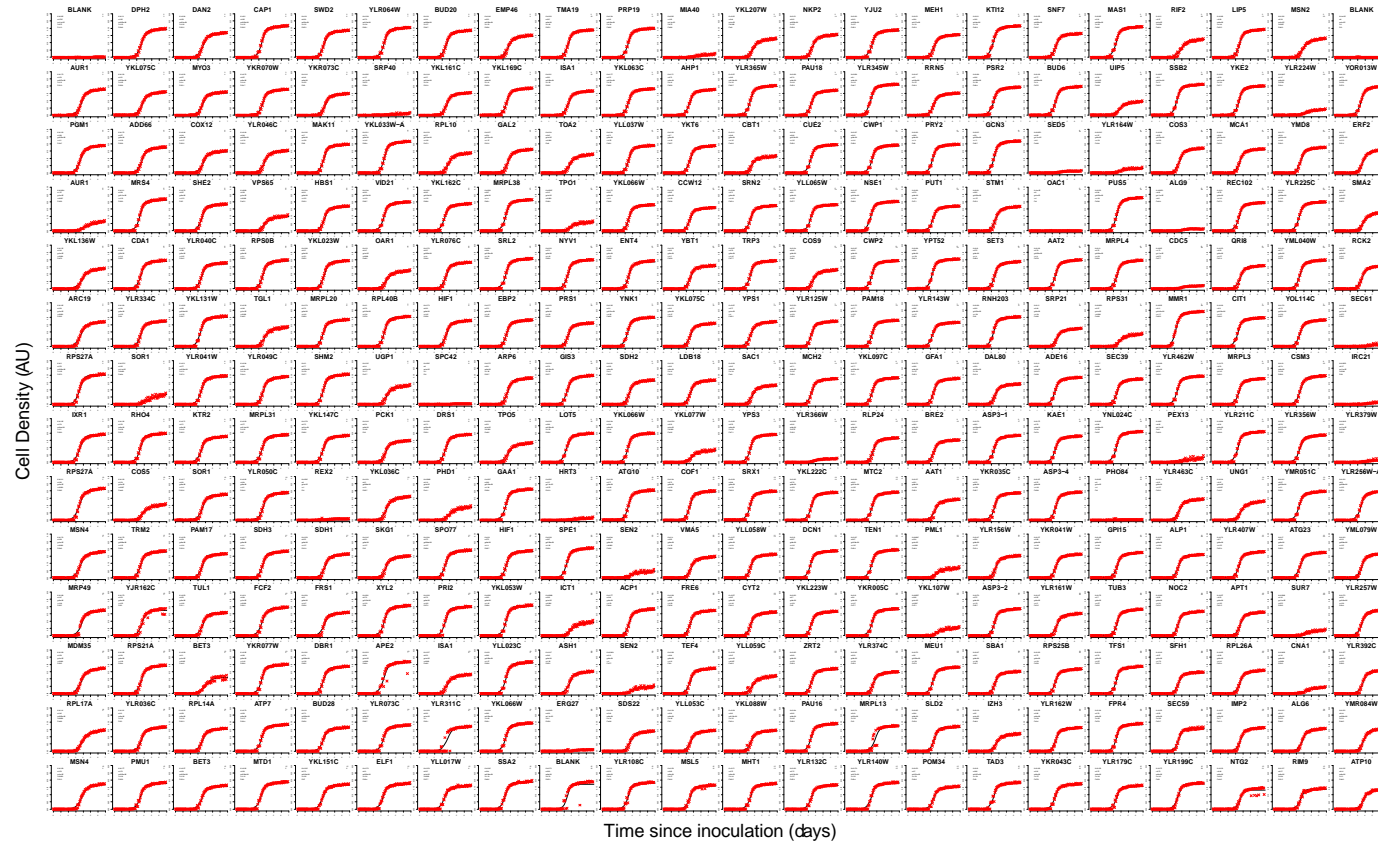


Quantitative Fitness Analysis

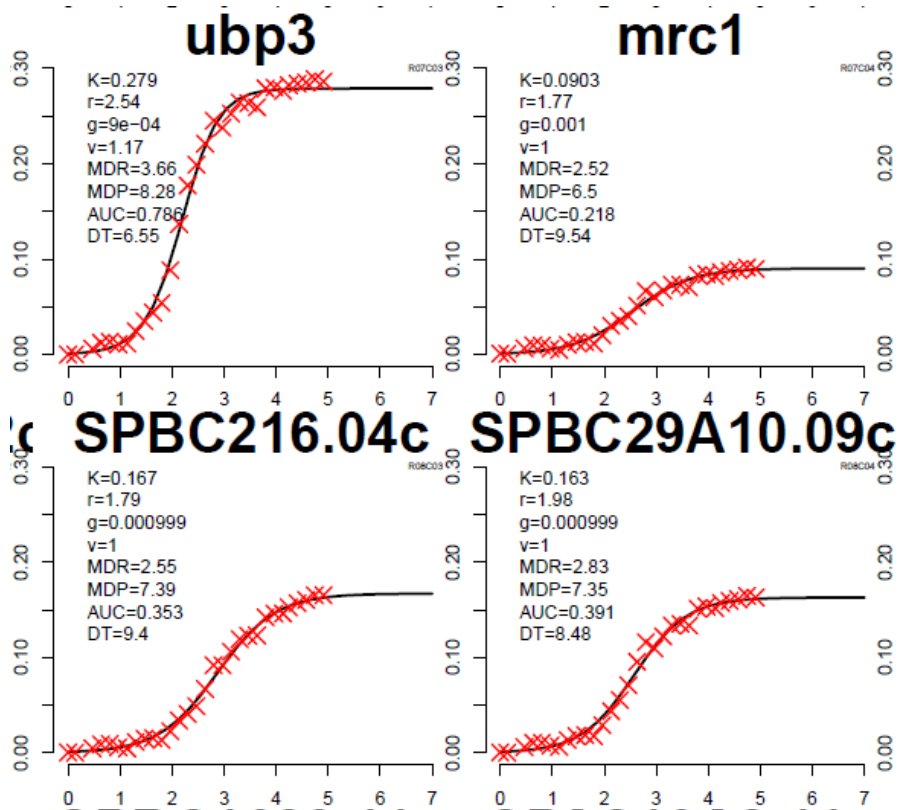


Quantitative Fitness Analysis

DLR00030513 Treatment: gal20 Medium: gal Plate: 9



Quantitative Fitness Analysis



R package version: 0.0-19

Treatment: 37

Medium: YES5_GH

Screen ID: QFA0089

Screen name: pot1-1

Libraries: pot1-1

Client: SID

User: SID

PI: DAL

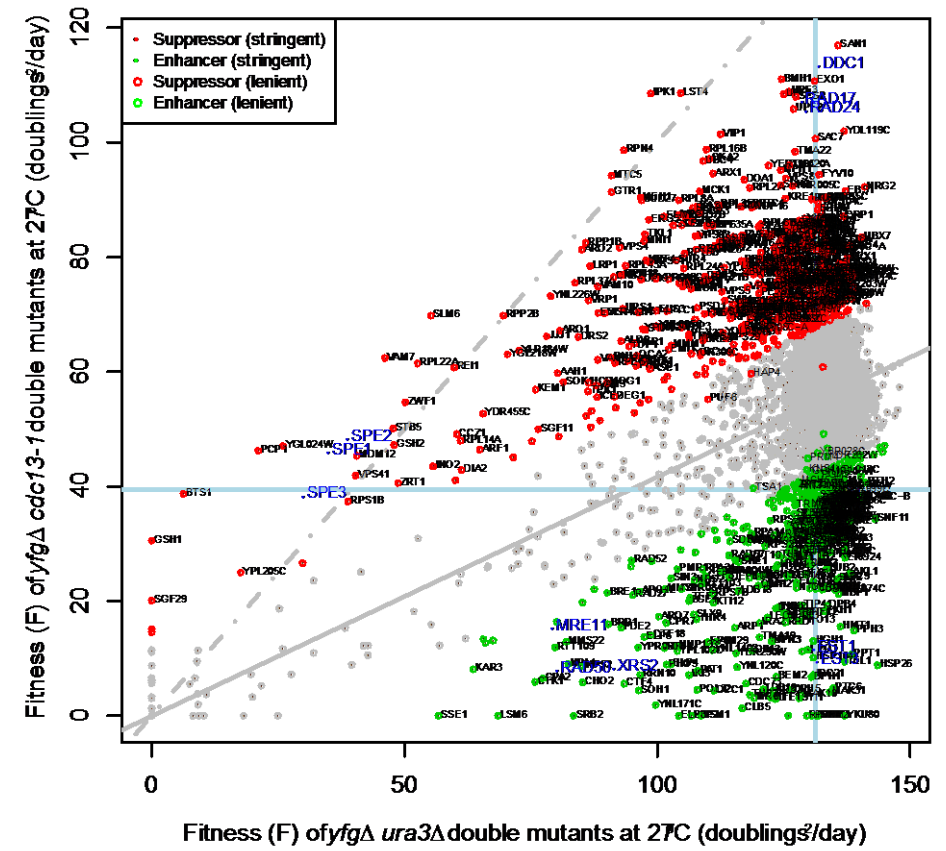
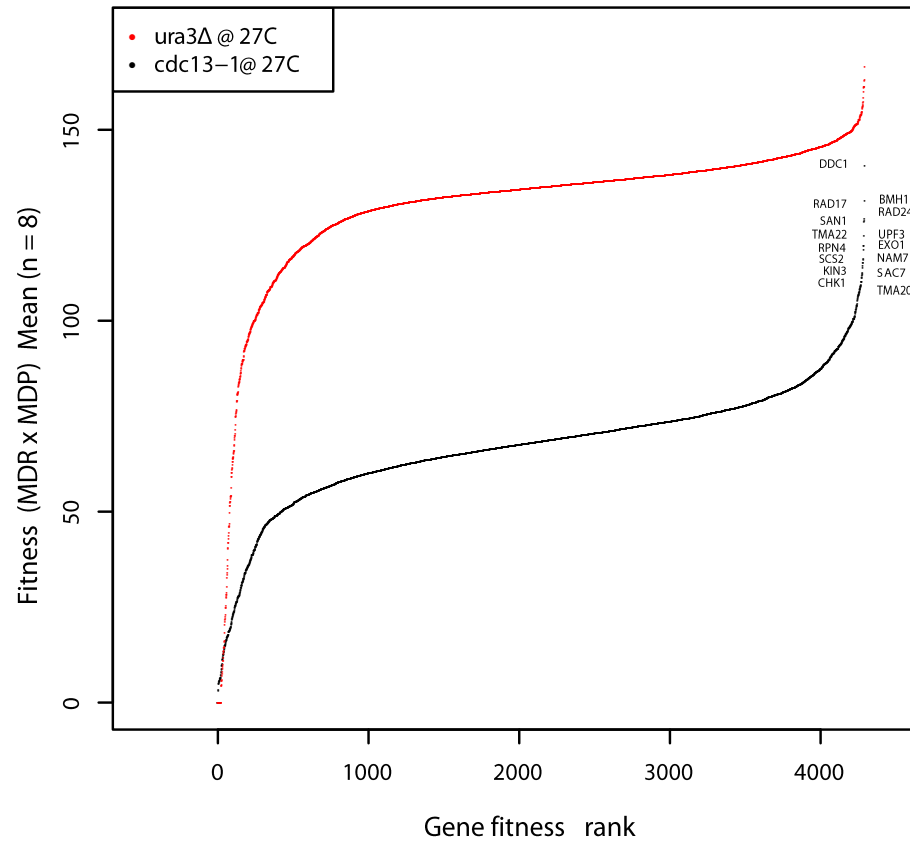
Date: 2014/04/11

Fitness definition: MDRMDP

Condition: YES5_GH

Gene	ORF	MedianFit	MeanFit	VarianceFit	NumRepeats	SEFit
SPAC1002.01	SPAC1002.01	45.67602071	45.55108874	136.5632446	4	5.843013876
pom34	SPAC1002.02	0	10.33812149	427.5070235	4	10.33812149
gls2	SPAC1002.03C	31.48131897	31.23084888	51.10339406	4	3.574331898
jmj2	SPAC1002.05C	39.17466695	36.10210929	180.6930891	4	6.721106478
bqt2	SPAC1002.06C	36.04907082	39.50708093	120.0299373	4	5.477908756
ats1	SPAC1002.07C	0	0	0	4	0
SPAC1002.12c	SPAC1002.12C	31.55007193	33.1592966	45.96544004	4	3.389890855
itt1	SPAC1002.14	26.91125138	24.27565593	114.9648816	4	5.361083883
urg2	SPAC1002.17C	18.81631727	19.47474406	4.967224907	4	1.114363597
urg3	SPAC1002.18	22.11026069	20.75929954	174.2771724	4	6.600703985
urg1	SPAC1002.19	46.23778821	45.37212432	3.520856264	4	0.938197243
SPAC1002.20	SPAC1002.20	35.8484572	33.2226362	54.75880496	4	3.699959627
psp3	SPAC1006.01	2.229726626	6.202746225	93.3917736	4	4.831970964
red1	SPAC1006.03C	0	0	0	4	0
mcp3	SPAC1006.04C	33.46108401	29.6629432	105.5012608	4	5.135690332
rgf2	SPAC1006.06	11.31532715	14.25079284	49.70679969	4	3.525152468
win1	SPAC1006.09	0	1.70233175	11.59173355	4	1.70233175
SPAC1039.02	SPAC1039.02	0	0	0	4	0
SPAC1039.03	SPAC1039.03	38.2425297	40.67746581	94.73005201	4	4.866468227
SPAC1039.04	SPAC1039.04	5.835202598	7.954314242	96.33651118	4	4.907558232
klf1	SPAC1039.05C	19.45426299	18.39563743	62.11796946	4	3.940747691
SPAC1039.06	SPAC1039.06	6.358457947	6.786677237	50.27796702	4	3.545347903
SPAC1039.08	SPAC1039.08	0	0	0	4	0

Data Analysis



Data Analysis – Alternatives

QFA tools are open source

- Colonyzer
<http://research.ncl.ac.uk/colonyzer/>
- QFA R Package
<http://research.ncl.ac.uk/qfa/>

R courses

- Colin Gillespie
<http://www.jumpingrivers.com//>

Bioinformatics Support Unit

- <http://bsu.ncl.ac.uk/support/>
- Based on 2nd floor Cookson

QFA: Quantitative Fitness Analysis of microbial cultures

Quantitative Fitness Analysis



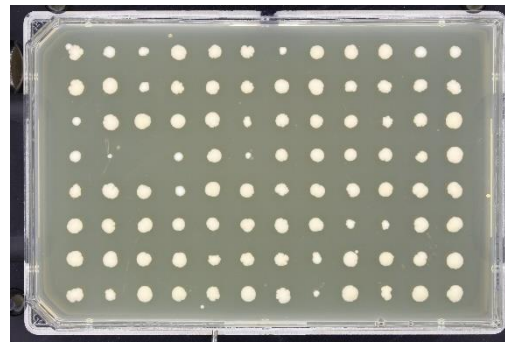
Random Colony Picking

Automated picking and arraying of random colonies

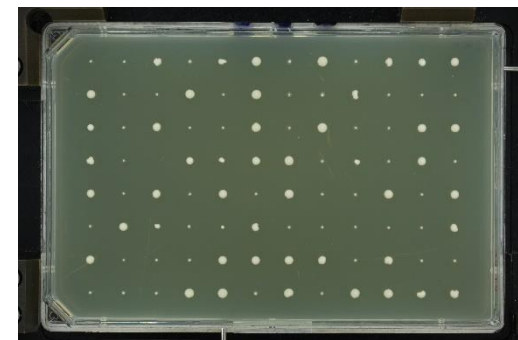
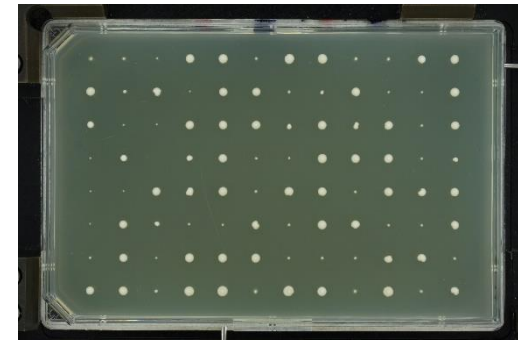
- Mutagenesis Screens
- Arraying plasmid libraries



→
Robotically
array colonies



↗
Test colonies
↘



MRC Technology Drug Library



MRCT Index library

- ~12,000 compounds selected as representatives of the full MRCT collection

MRCT kinase library

- ~6,700 compounds predicted to inhibit kinases

MRCT natural product library

- ~4,000 purified novel natural products from plants or fungi

FDA approved drugs library

- ~1,400 compounds

ABOUT US

MRC Technology is an independent life science medical research charity committed to drastically improving positive patient outcomes everywhere.

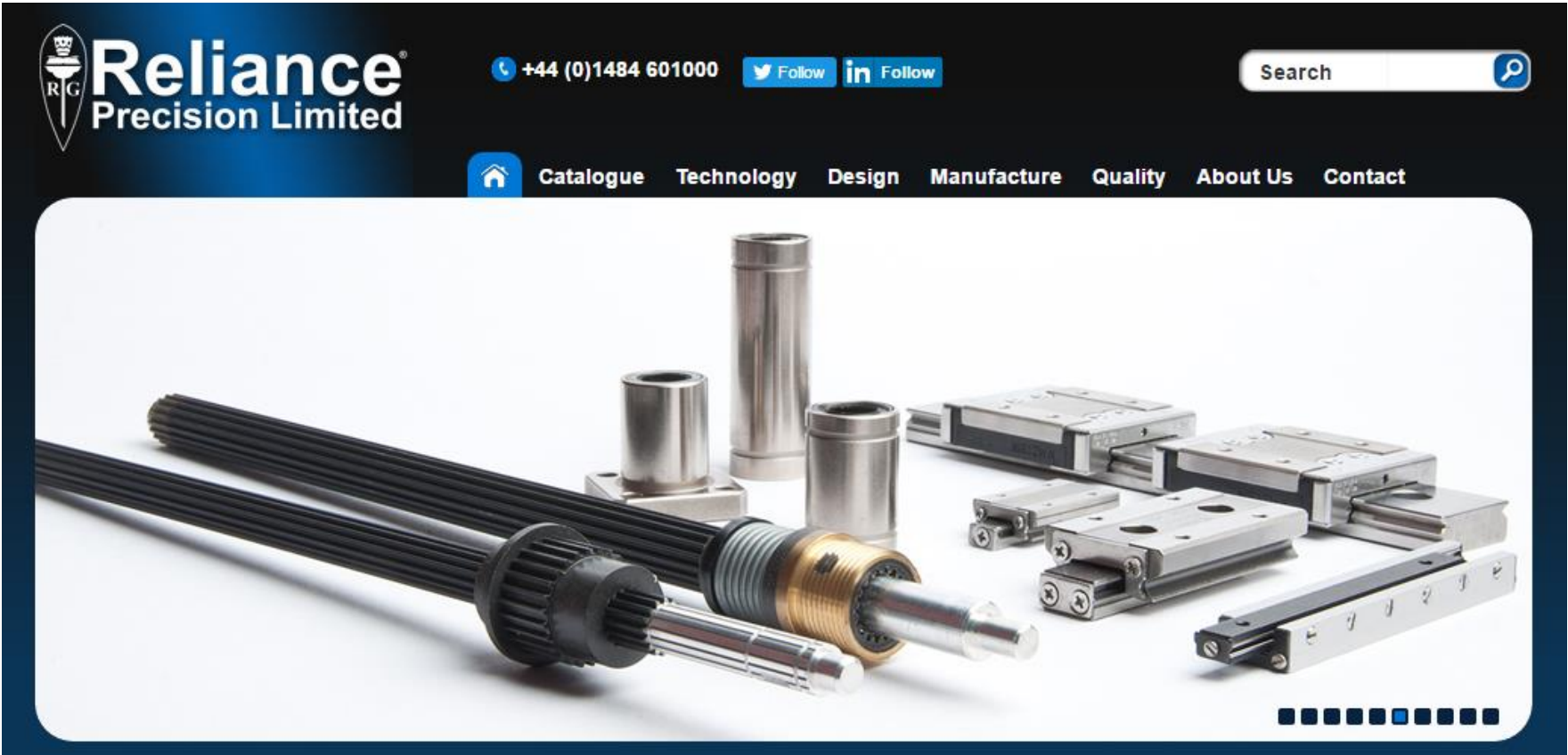
As a champion for human health, MRC Technology partners with academic, biotechnology, pharmaceutical, and charity organisations to move promising medical research forward into viable and accessible patient treatments.

Our people combine commercialisation and technology transfer/IP management skills with diagnostic and drug discovery expertise, specialising in small molecule and therapeutic antibodies. MRC Technology projects have led to the approved drugs Tysabri®, Actemra®, Entyvio® and Keytruda®, changing the lives of countless patients by harnessing the potential of science.

Costs

	Cost £/Hour Standard Robotic Systems	Cost £/Day Automated Imaging systems
With technical assistance	35.81	24.25
Without technical assistance	10.99	7.20

New Robotics



Acknowledgements

Dave Lydall

Neil Perkins

Martin Cox

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HTSF Website

<http://research.ncl.ac.uk/bioHT/>