Identification de molécules « first in class » : le High Content Screening



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What is HCS ?

Images contain information that can be quantified!



From Images to Numbers



Calculate properties

Rel spot intensity: 0.11 Number of spots: 29 Total spot area: 39 µm² Cyto area: 193 µm² Spot area/Cyto area: 0.20



From Numbers to Knowledge



Chloroquine treatment leads to autophagosome accumulation





Phenotypic Drug Discovery Identifies New Drug Targets

HCS: Phenotypic drug discovery



- Hypothesis-free
- Identification of compounds causing a desirable change in phenotype in a disease relevant cell or animal model

HTS: Target-based drug discovery

Target

- Hypothesis-driven
- Identification of compounds modulating the activity of a biological target
- Requires the identification of a druggable target
- Often starts with large biochemical screens against the purified target



Gain More Information From Your Cellular Samples

High Throughput Screening



1 well – one readout

6 Complementary tools for drug discovery

High-Content Screening



1 well – many readouts, phenotypic information



The phenotypic approach

Phenotypic assay in a disease relevant cellular model "Phenotypic fingerprint"

Multi-parametric analysis of phenotypic changes



Quantify as many features as possible!



"Feature Selection"

Select features with most descriptive power using machine learning technology

Identify "Hits" Compounds, siRNAs, molecular targets







Exemple : Cambridge Biomedical Research Center



Microplaque CellCarrier Ultra 96 puits









50 um





Exemple : Cambridge Biomedical Research Center



Exemple : Cambridge Biomedical Research Center

Exemple : Zoffmann et al. (Roche)

E. coli

Membrane DNA membrane permeability

Globomycin

Mecillinam

Ceftriaxone

Levofloxacin

Colistin

Ciprofloxacin

Polymyxin B

Doxycycline

Chloramphenicol

(2019) 9:5013 | https://doi.org/10.1038/s41598-019-39387-9

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PerkinElmer Instruments for HCS

Opera Phenix Plus

Operetta CLS

Choice of Magnification

14 To fit different applications, plates and needs

20x W NA 1.0

- 4.7MPx sCMOS camera
- Air and Water immersion objectives
- Widefield and confocal imaging
- Brightfield and digital phase imaging

- Fast, high resolution x,y-table and single lens z-drive
- 6 position objective turret
- Sample based flatfield correction
- Easy to use Harmony software
- Automation ready

Add More Cameras for Increased Speed

Cam 1

Cam 2

Cam 3

Cam/4

Operetta CLS Sequential acquisition (1 camera)

Opera Phenix Plus Simultaneous acquisition (2 or 4 cameras)

16 Multi-camera systems speed up experiments

Intelligent Aquisition – Spheroids in Matrigel

PreciScan (XY & Z)

Pre-scan organoids with low magnification z-stack

Identify xy and z positions of organoids in matrigel

Re-scan – Measurement (63x)

Re-scan individual organoids with high magnification z-stack

Image Analysis Workflow- Constructed From Building Blocks

Segment image to identify objects

Refine regions of interest

Classify define and select specific cellular populations

Multiple Ways for 3D visualization

19 and Maximum Intensity Projection

3D Volumetric Analysis of Luminal Spaces Inside Cysts or Organoids

Define hollow space

ValueCalculateSpecific properties for culuationSpecific properties for culuation		
yst Centroid X in	Cyst Centroid Y	Cyst Centroid Z in
lmage [µm]	in Image [µm]	lmage [µm]
-117.0	13.1	47.0
86.5	-38.5	66.6
-1./	-22.5	58.0
Cyst Volume [µm³]	Cyst Surface Area [µm ²]	Cyst Sphericity
168879	17877	0.83
217712	21292	0.82
254947	23239	0.84
Lumen Volume [µm³]	Number of Nuclei- per Cyst	Cell Volume [µm³] - Mean per Cyst
33415	139	813
63350	130	1080
59776		1000
33770	179	922

explorer[™] G3 workstation Automate custom workflows and increase productivity and reproducibility

CellCarrier[™] imaging plates Optimal clarity and fast autofocusing from excellent flatness of the plate bottom

Opera Phenix Plus

Maximal speed and sensitivity for 3D assays

Operetta CLS

Sensitivity and flexibility for all your biology

Image Analysis

Harmony Software for robust description of cellular phenotypes with texture and STAR morphology and showing results

Scalable Screening Solutions

Opera Phenix® with plate::handler™ Flex for Fixed Cell

For inscreasing needs

HELPING LABS DO MORE with LESS

IMPROVE EFFICIENCY

INCREASE PRODUCTIVITY

- Enhanced walk-away time
- Continuously processing
- Easy set-up

REDUCE HANDS-ON TIME

COMPLIANCE

STANDARDIZATION

explorer[™] G3 workstation iX20

(EXAMPLE CONFIGURATION with optional enclosure)

J SignalsScreening

Dimensionality reduction, feature selection and phenotypic profiling Plate values Feature overview Exploration of data structure Class definition Feature selection and classification × Hit detection **Feature Ranking & Hit Selection** Feature importance scores Selected features Feature Feature rank hc_Nuclei - Cytoplasm Tubulin SER Edg... 1.00

Unsupervised **Machine Learning**

How to upscale / automate?

Liquid Handling

Automate liquid handling

High Content Imaging Run your HCS system day and night

Run image analysis synchronized with image acquisition

Secondary Data Analysis

Use dedicated screening software for automated and standardized data analysis

Thank You

Training users in proper use of the Opera Phenix system has been a simple process. Users who have some HCS experience have been trained in-house and quickly took to visualization and quantification with the new system. Less experienced users have been able to collect their own data with less than an hour of training, and with assistance can now process their data to achieve automated quantification.

Steve Bagley

Head of Imaging & Cytometry, Cancer Research UK, Manchester Institute, The University of Manchester

PerkinElmer HCS in Europe

PerkinElmer

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Integrations

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- Designed to Run24/7
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Throughput

Detection & Imaging Systems to Analyze Models of Increasing Complexity

