NANOSCIENCE 2009
Lichtenwalde

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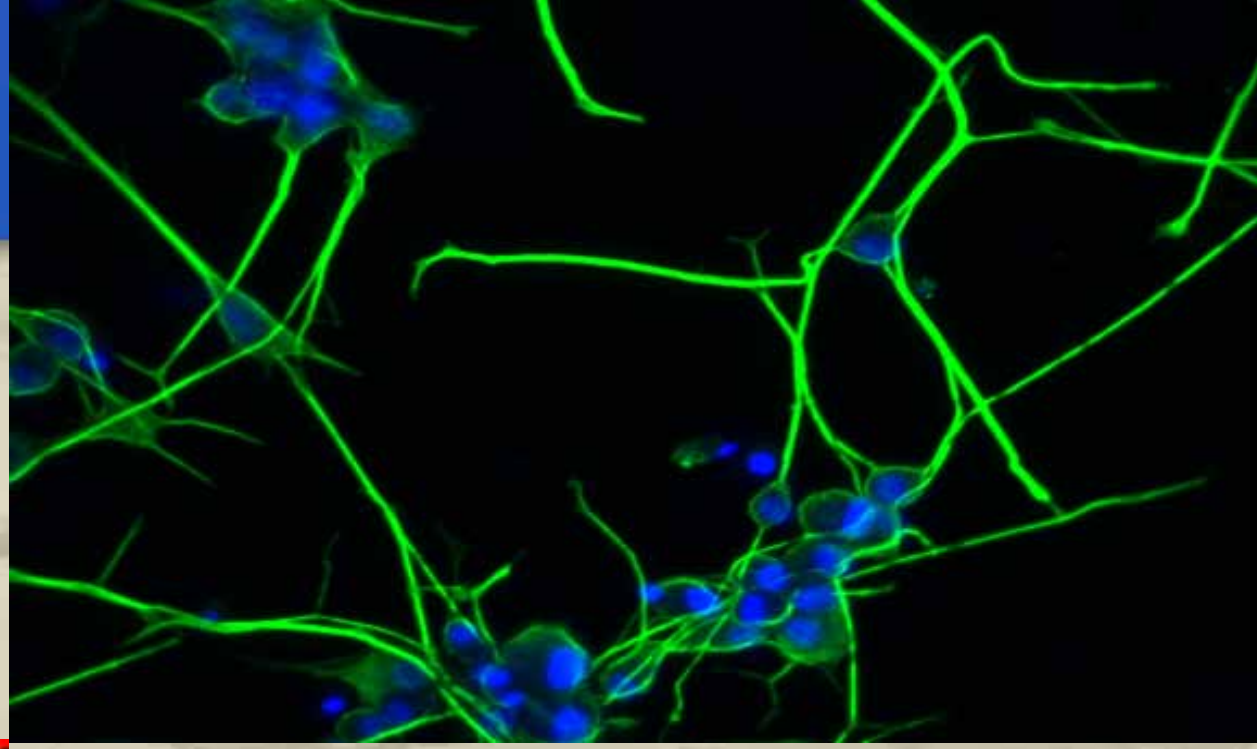
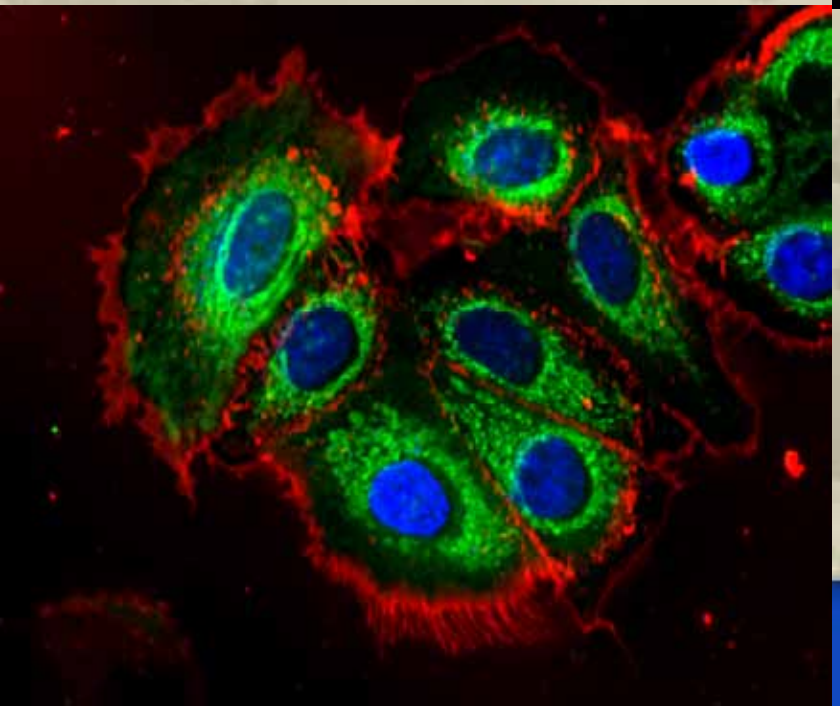
BD Biosciences
European Technology Center
Allschwil, Switzerland





Development of cellular assays for automated microscopy to measure multiple parameters by single cell high content analysis

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BD Biosciences

BD Biosciences

BD Cell Analysis

BD Flow Cytometry

BD Bioimaging

BD Pharmingen

BD Discovery Labware

- High content imaging systems
- Analysis software
- Bioimaging reagents
- Cellular Reagents

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Instrumentation has Enabled the Scaling and Automation of Biology

Increasing Complexity

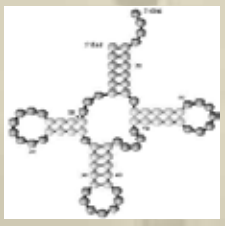
Genome (DNA)

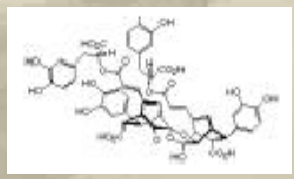
Transcriptome (RNA)

Proteome

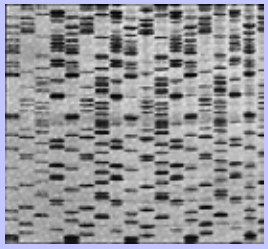
Metabolome Physiome

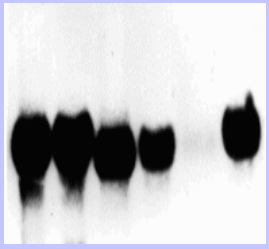
Cell/Tissue/System/Organism

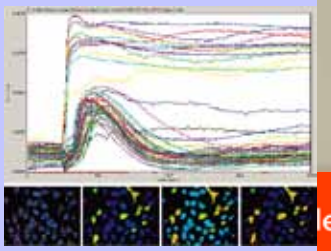




Scaling

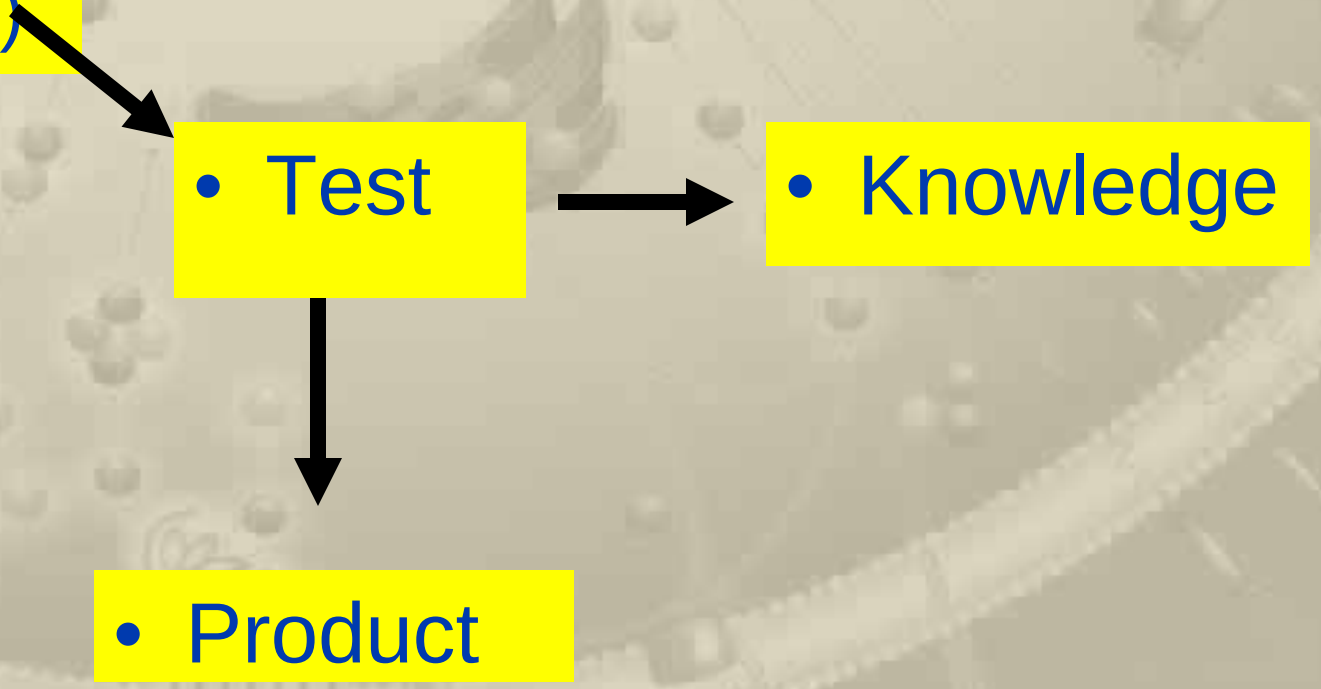






The Problem we want to address:

- Biological and medical applications (e.g.Nanoscience)



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The Problem we want to address:

- Test in Biological models :
  - Healthy organism
  - Disease models

à Microscopy -> Pictures -> Data

- Problem:

Time consuming repetetive work

- Solution:

automated microscopy and High Content Analysis

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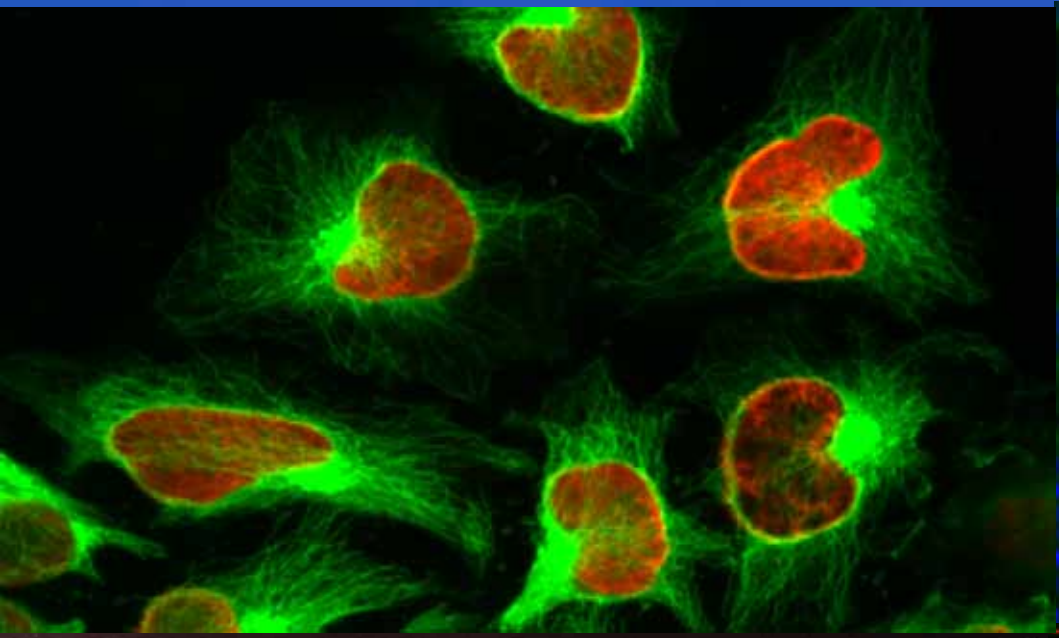
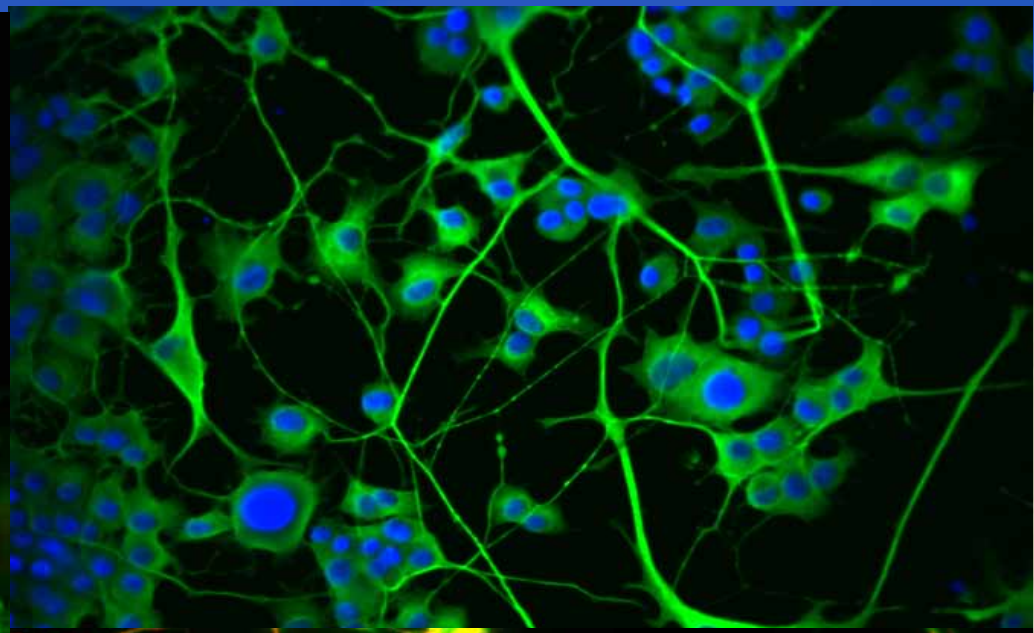
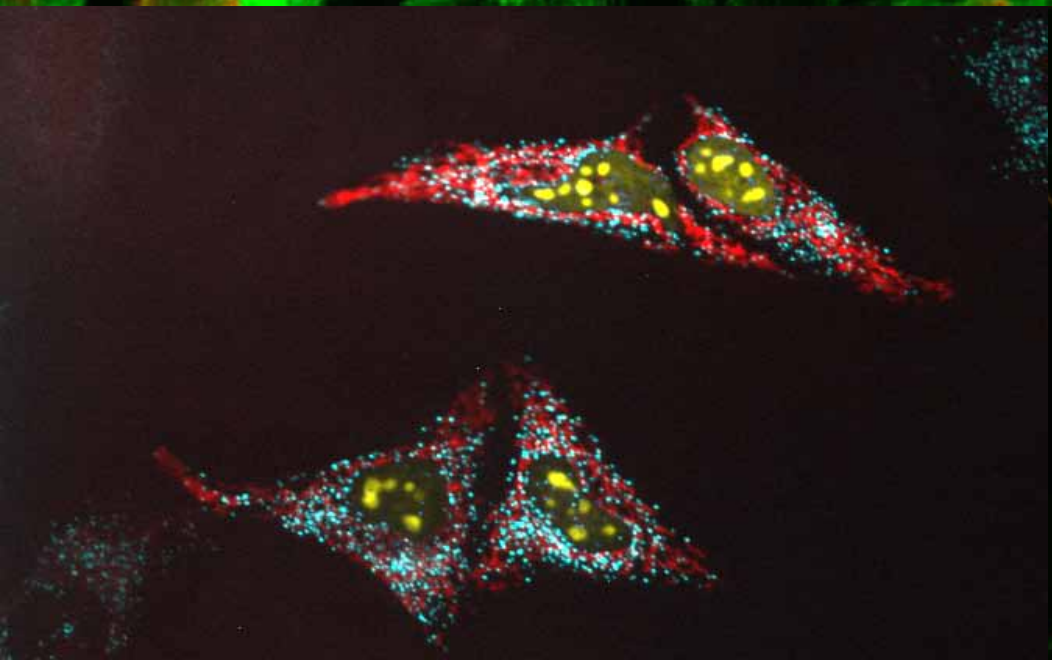
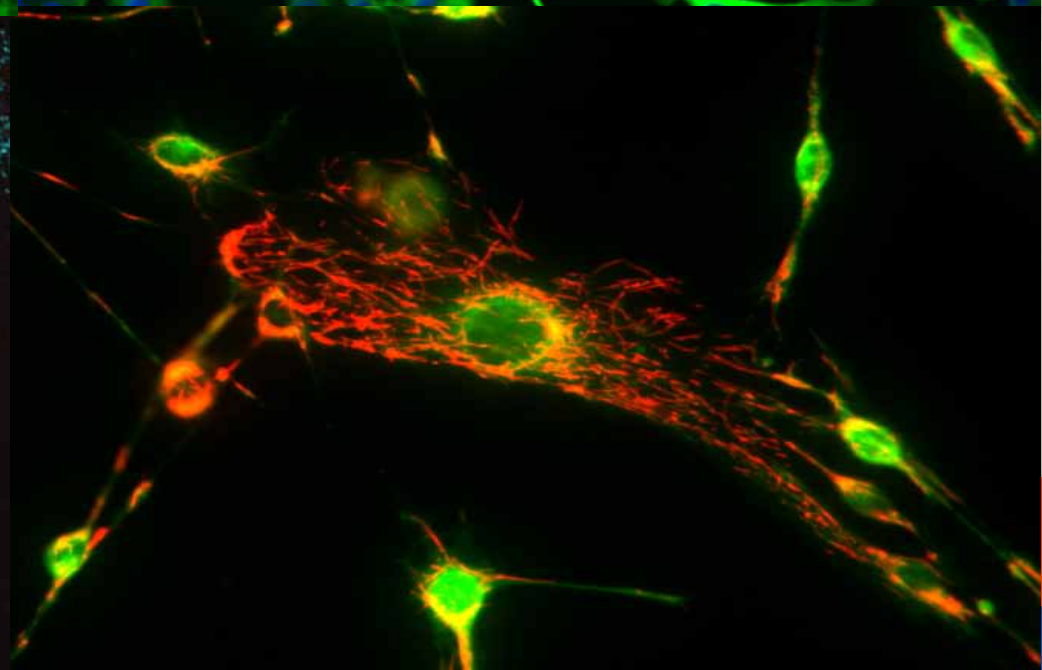
Goal: to automatically acquire images of multiplexed assays and…

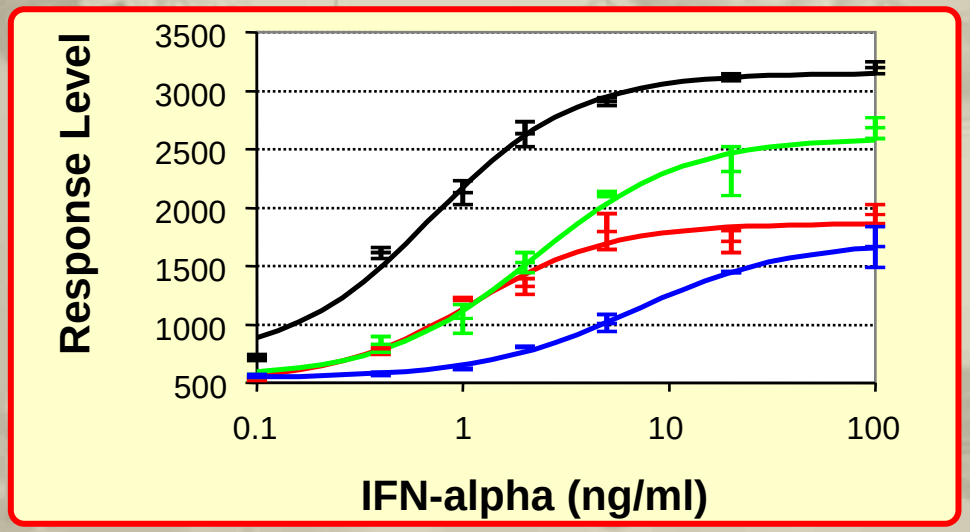


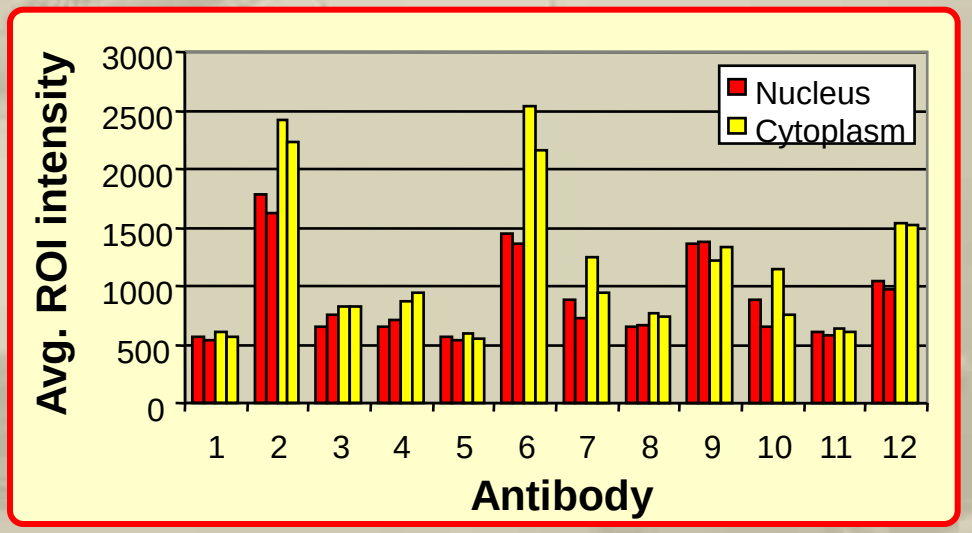


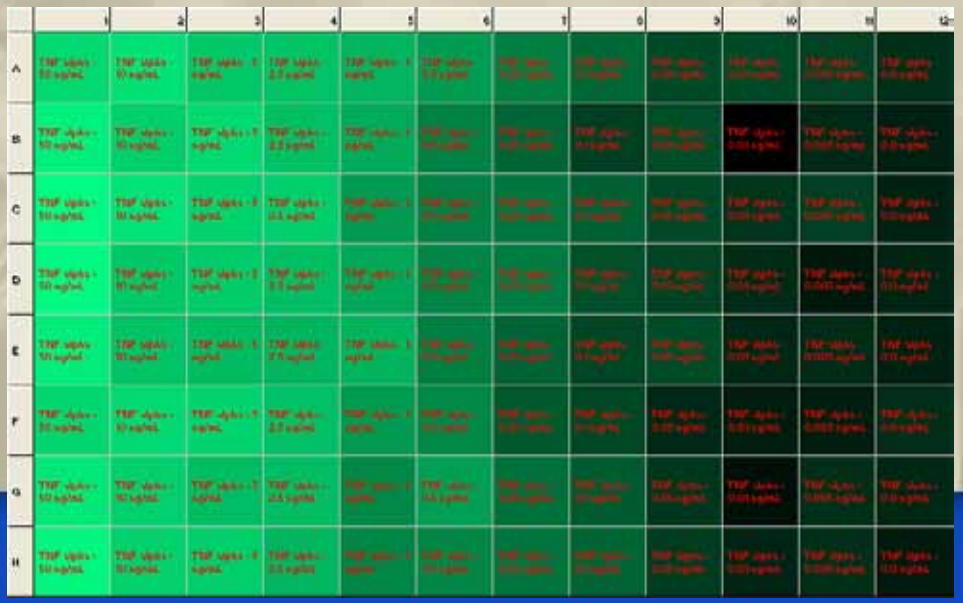




…to turn such images into meaningful data





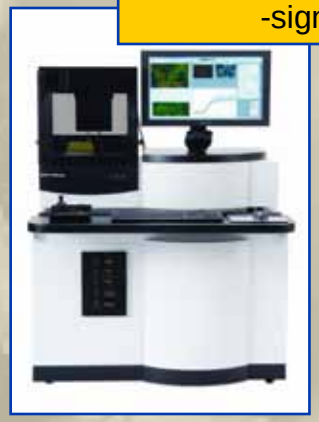


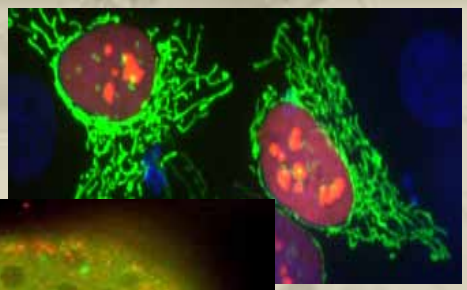
	A	B	C	D	E	F	G	H	I	J	K
1											
2	WELL DA	Report(Report_1)									
3	DATA SOI	Fluo_4___Plate_Map			Fluo_4_RoiSummary			ROIs			
4	CONSTRA	NONE									
5											
6		Dose		ROI Count	ROI Count		Verticies	Delta	Delta		
7	Well ID	Label	Dose	Before Cor	After Corrs	% of Total	Average	80% Lowe	80% Uppe	Std Dev	Std Err
8	A001	5	5E-09	76	76	100	120.7237	61.72368	106.2763	96.33367	11.12365
9	A002	5	5E-09	112	112	100	146.3571	82.35714	123.6429	132.0132	12.53014
10	A003	5	5E-09	77	77	100	97.24675	38.24675	40.75325	46.20639	5.300236
11	A004	5	5E-09	97	97	100	127.3814	60.30144	112.6186	99.76279	10.102
12	A005	10	1E-08	99	99	100	200.101	133.101	88.89899	517.8083	52.30654
13	A006	10	1E-08	108	108	100	116.6574	54.65741	102.3426	77.21268	7.464431
14	A007	10	1E-08	123	123	100	115.4797	53.47967	105.5203	60.44363	5.472309
15	A008	10	1E-08	131	131	100	138.7328	75.73282	96.26718	84.2875	7.392502
16	A009	50	5E-08	106	106	100	108.3019	47.30189	67.69811	65.0335	6.346619
17	A010	50	5E-08	140	140	100	118.2357	56.23571	116.7643	83.05607	7.044724
18	A011	50	5E-08	140	140	100	168.8714	99.87143	135.1286	178.0452	15.1016

The High Content Imaging Process and BD Bioimaging Portfolio

Bioimaging- Certified Reagents
Biol. Models and Kits

BD Pathway Bioimager 855 +435
-optimized image quality
-stage precision
-versatile autofocus
-signal/noise stability + speed





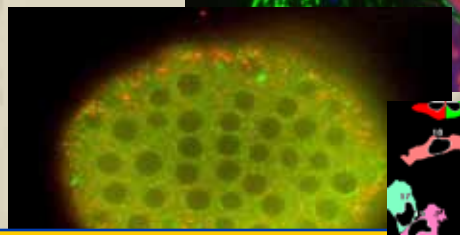
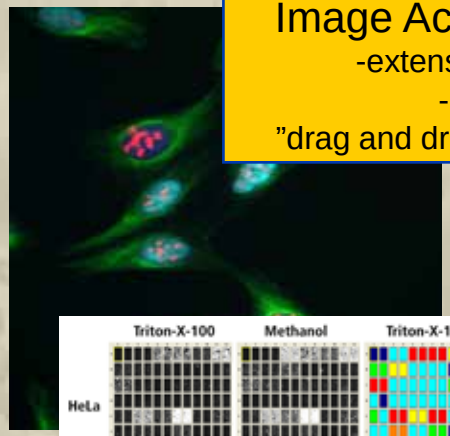
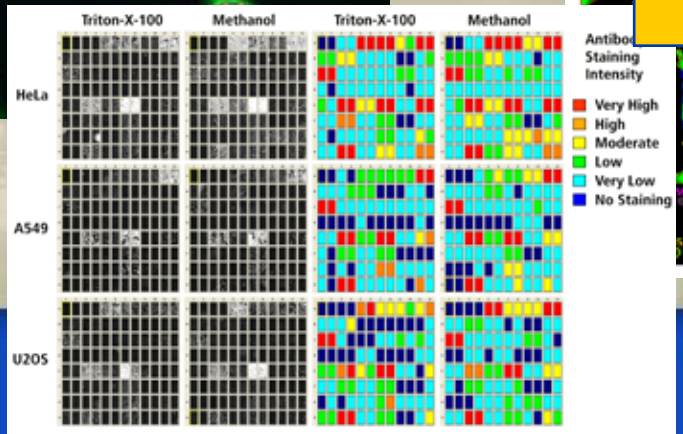
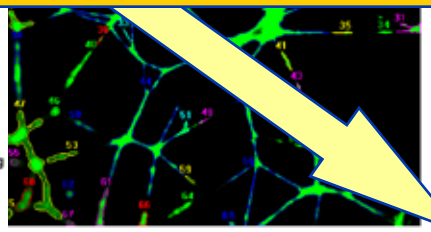


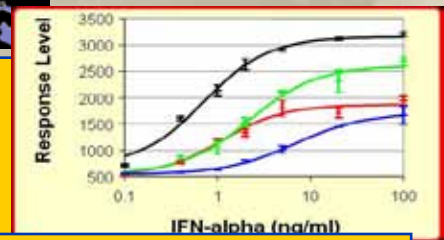
Image Acquisition software
-extensive normalisation
-multiplexing
"drag and drop" assay development

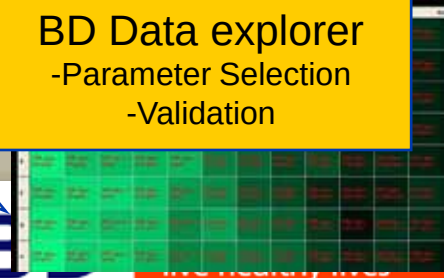




HCA ImageAnalysis
-multiple algorithms
-automated object finding







BD Data explorer
-Parameter Selection
-Validation

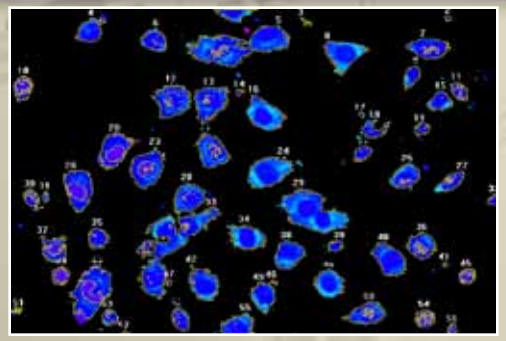
Z'>0.5

Secure
Hit Identification

High Content Image Analysis

- 1.Step: Automatic **acquisition** of images of cultures, tissues or whole organisms.
- 2.Step Heart of automated analysis is **object detection** = finding the single cells or other structures automatically in the image
- 3.Step: Extraction of information from these objects: intensities, distribution and morphology of multiple fluorescent labels

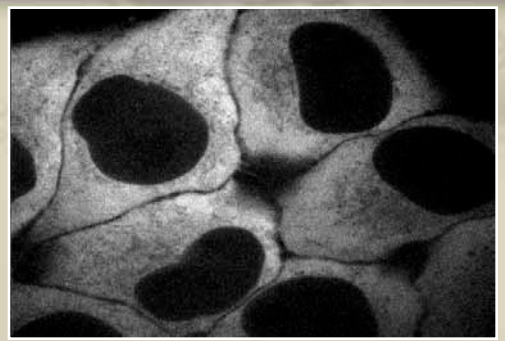
BD Pathway™ Bioimager: 1. Assays
General Assay Categories: functional classification



Fluorescence intensity change

Examples:
- Calcium flux
- Phosphorylation
- Protein expression
- Protein degradation
- Image cytometry

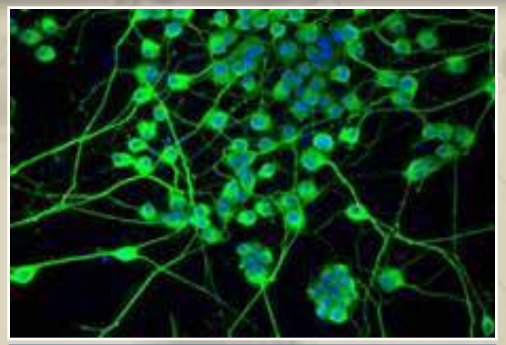
1



Fluorescence distribution

Examples:
- Cytoplasm to nucleus (e.g. NFkB)
- Cytoplasm to plasma membrane (e.g. PKCa)
- Plasma membrane to organelle (e.g. Transfluor GPCR)
- Receptor internalization
- Protein co-localization

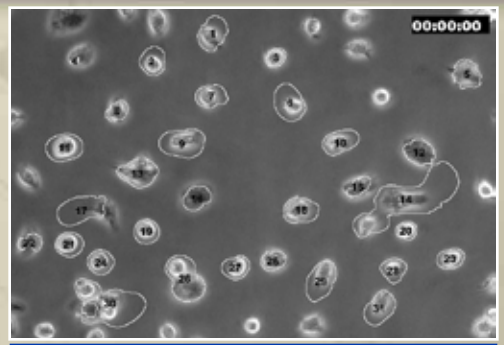
2



Morphology

Examples:
- Neurite outgrowth
- Angiogenesis
- Cell differentiation
- Apoptosis

3

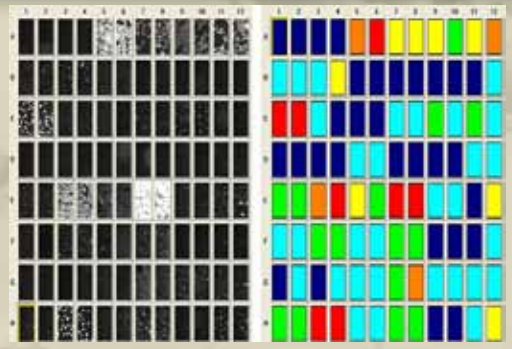


Movement

Examples:
- Chemotaxis/migration
- Wound healing
- Metastasis/invasion
- Migration, Tracking
- Cell division
- Rounding

4

BD Pathway™ Bioimagers: 2. Assays
General Assay classification based on protocol

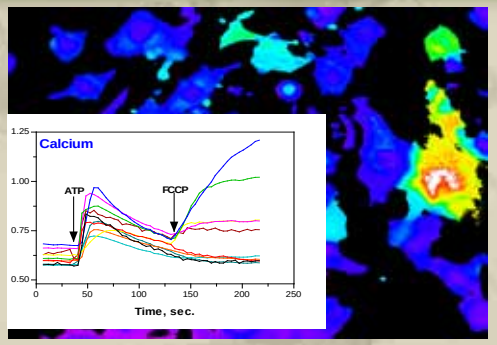


Endpoint Assays

Examples:

- Chemotaxis/migration
- Wound healing
- Metastasis/invasion
- Translocation
- Protein expression
- Whole organism (e.g.Zebrafish)
- ADME/Tox.
- Angiogenesis
- Neurite Outgrowth

1



Kinetic Assays

Examples:

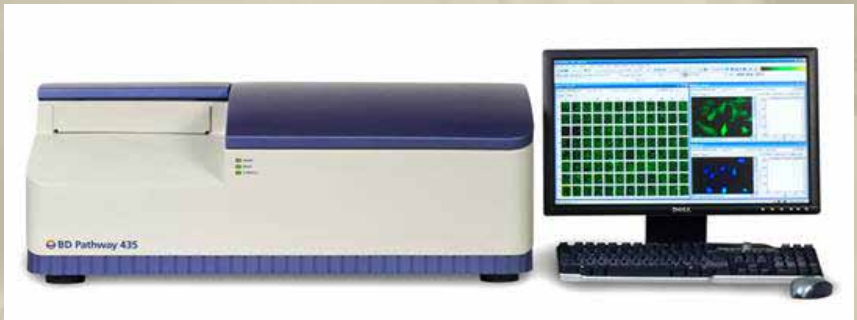
- Calcium flux
- FRET
- Phosphorylation
- Protein expression
- Protein degradation
- Image cytometry
- Tracking
- Differentiation
- Membrane potential

2

The BD Bioimaging Portfolio

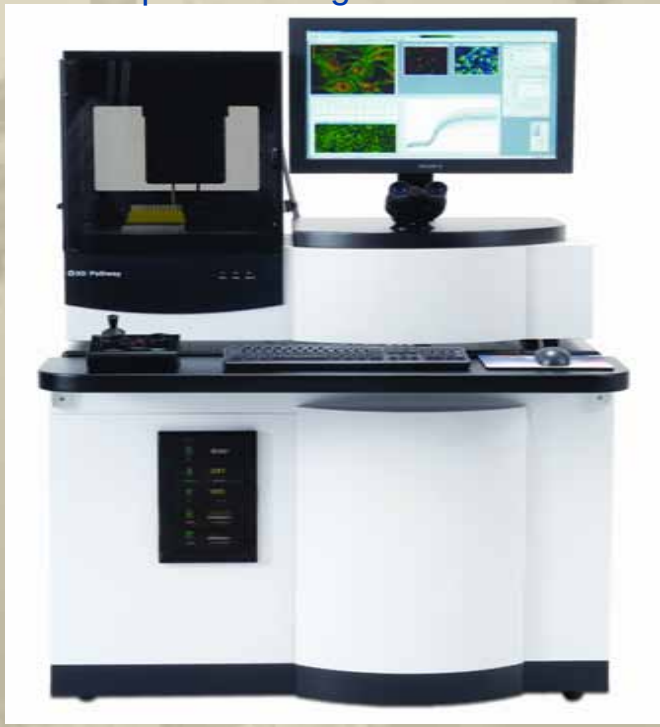
Pathway 435

High-performance, full spectrum [confocal] automated imager with powerful image analysis software



Pathway 855

The ultimate flexible live-cell kinetic system with environmental control and liquid handling



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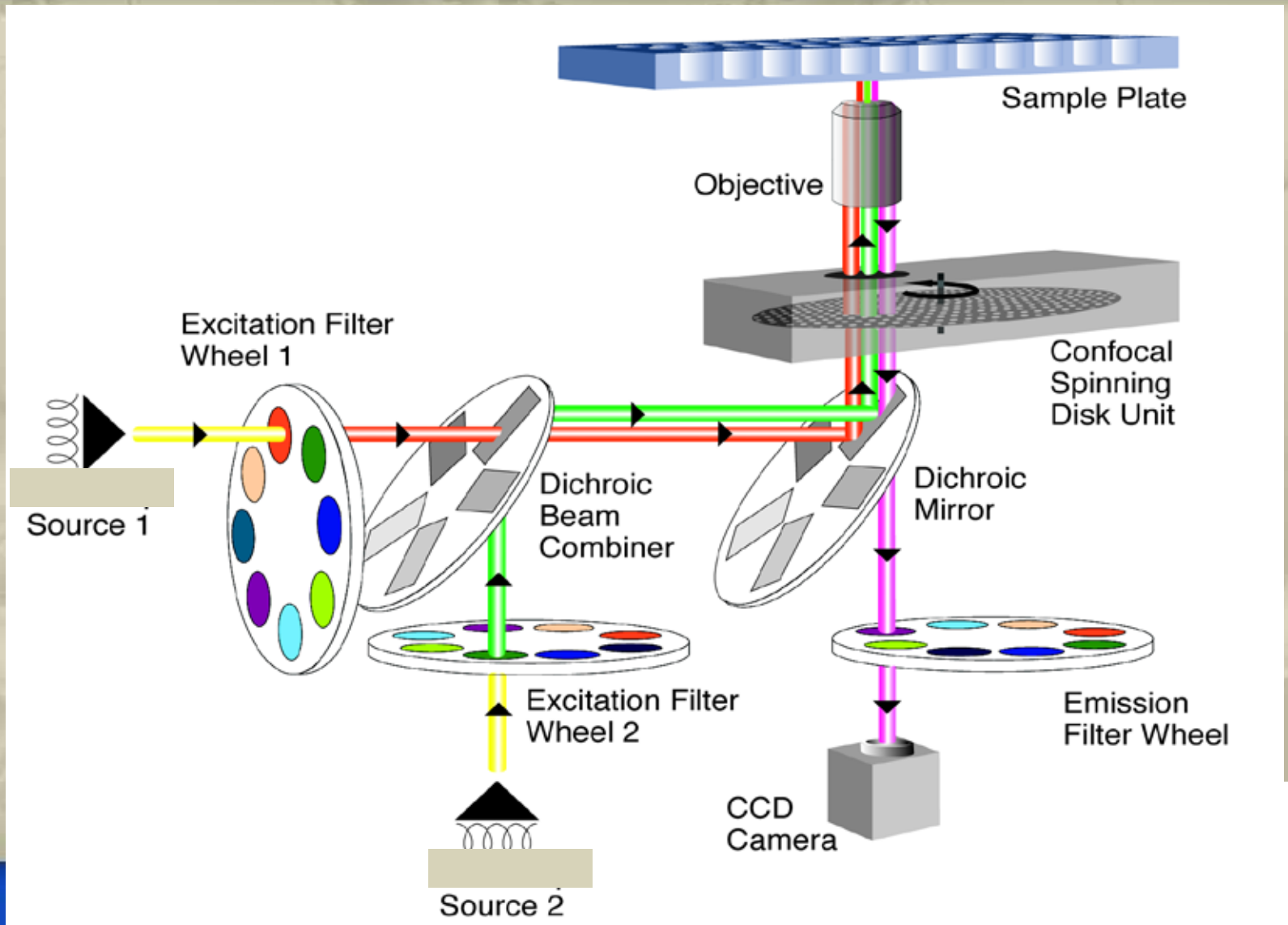
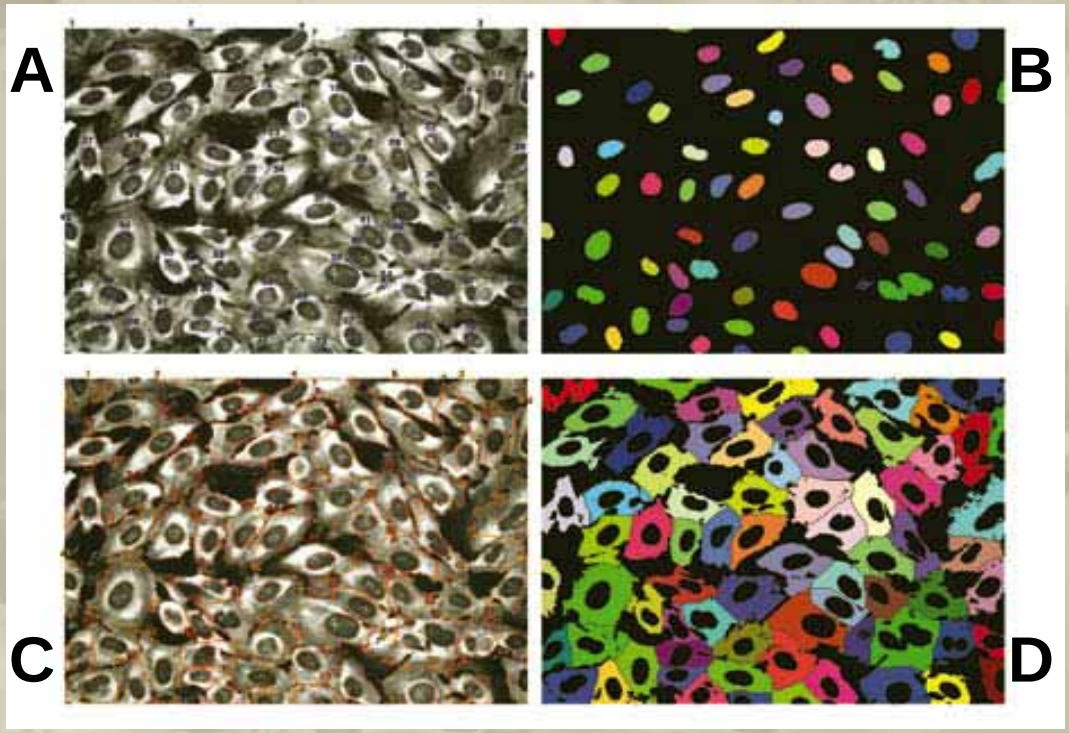
BD Pathway™ 855 Bioimager: 1. Hardware



3. Examples for Applications

3.1. Endpoint assays

Assay Examples:
Screening Antibodies for their suitability to use them in Cell Based Assays



Examples of segmentation of the fluorescent signal into specific regions of interest (ROIs), corresponding to the nucleus (A) and cytoplasm (C).

Panels B and D show the corresponding segmentation masks.

Representative images from a purified antibody screen using FITC goat anti-mouse IgG second step reagent

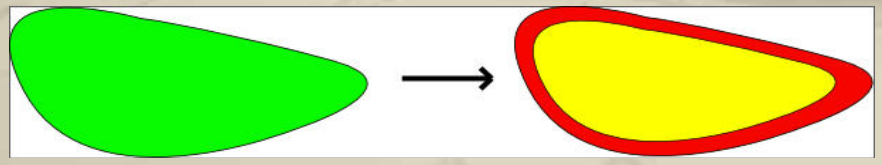
Bioimaging Certified Reagents

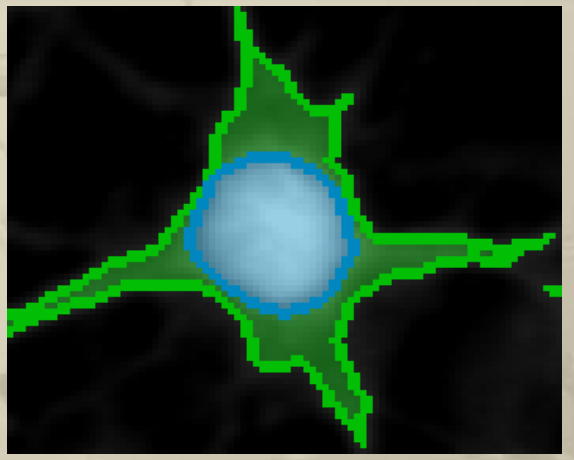
5 Product Groups

- Unlabeled Primary Antibodies – hundreds of specificities in the areas of cell cycle, apoptosis, signal transduction, cancer, stem cells, neurobiology…
- Directly Conjugated Antibodies – growing subset of Abs
  - Many specificities available in 2-3 colors
  - Enables high-level multiplexing, reduces processing time and steps
- Kits - Pre-qualified reagents and protocols
- Fluorescent Protein Organelle and Cell Structure Vectors
- Imaging Plates
  - Thin bottom multi-well, plates for optimal image quality

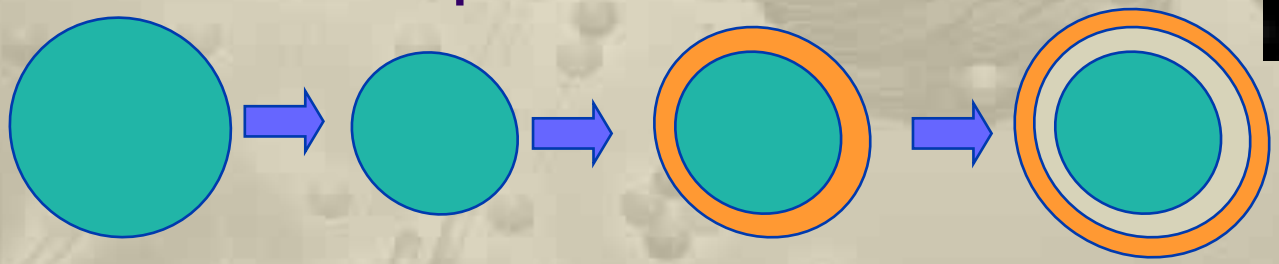
BD Pathway 855: Examples for Applications
AttoVision 1.6 new Segmentation Options

- **Membrane Segmentation**

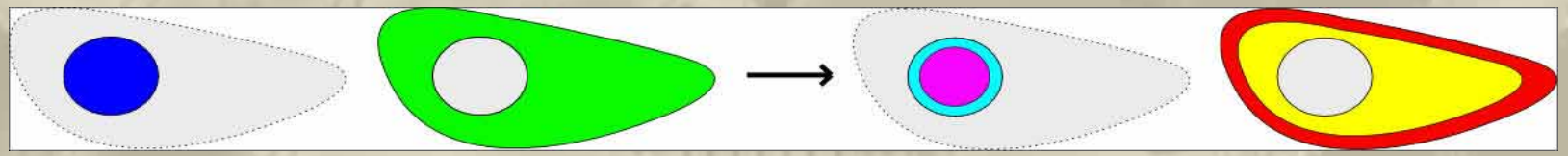




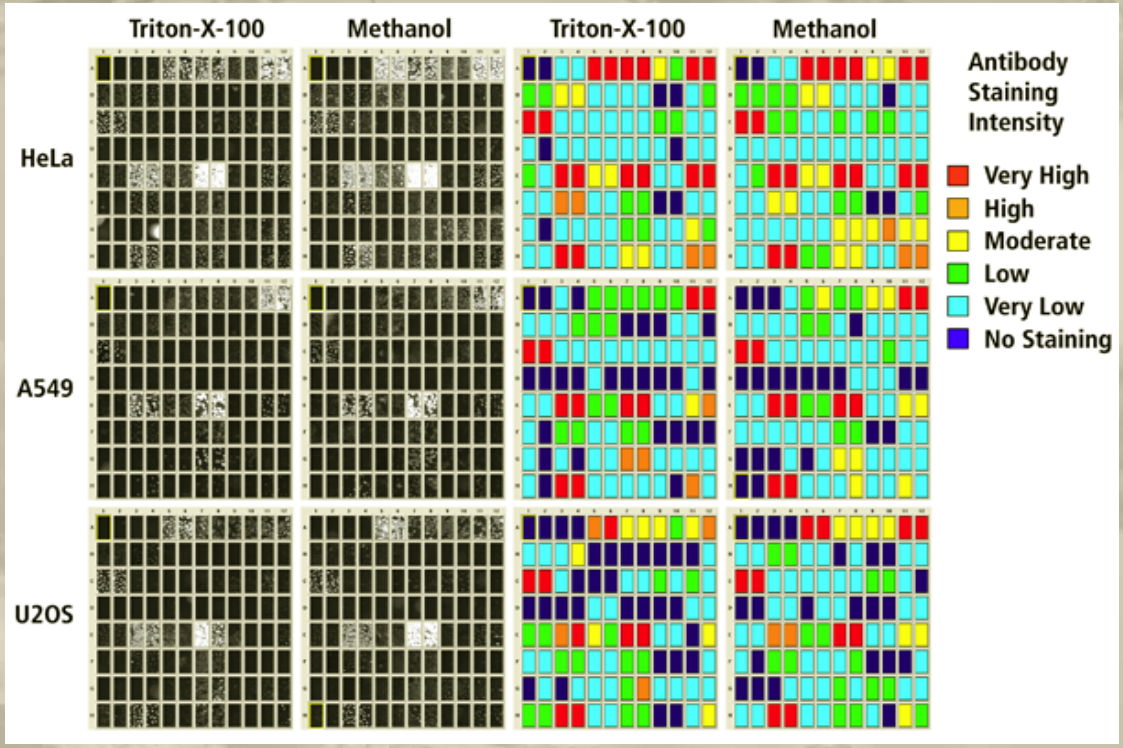
- **Translocation Gap**



- **Membrane Segmentation + Cytoplasm + Nucleus**



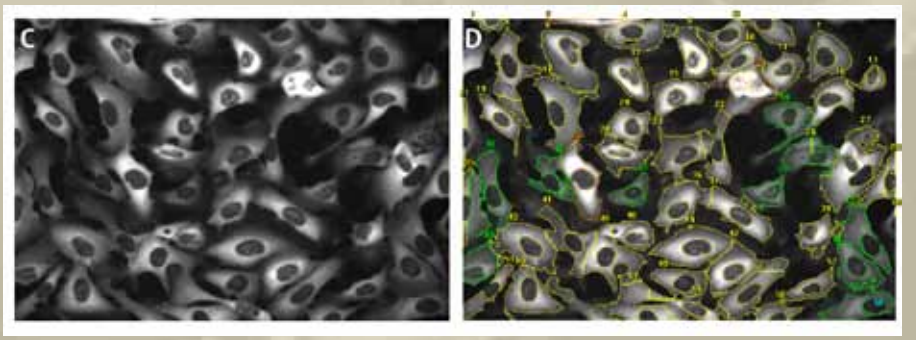
Application Examples:
Screening Antibodies for use in Cell Based Assays



Representative data from a 47 antibody screen in six 96 well plates

Full automation allows rapid evaluation of:

- **Multiple Antibodies**
- **Various fixation & permeabilization methods**
- **Antibody dilution**
- **Saving large amounts of time & increasing productivity**



Antibody screening data classified for intensity of cytoplasmic staining using 6 user-defined intensity levels. Cells are color-coded by the software based on their classification level, wells are classified based on the percentage of cells – a “moderate” well is shown

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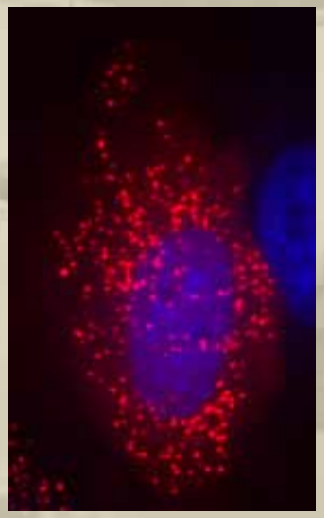
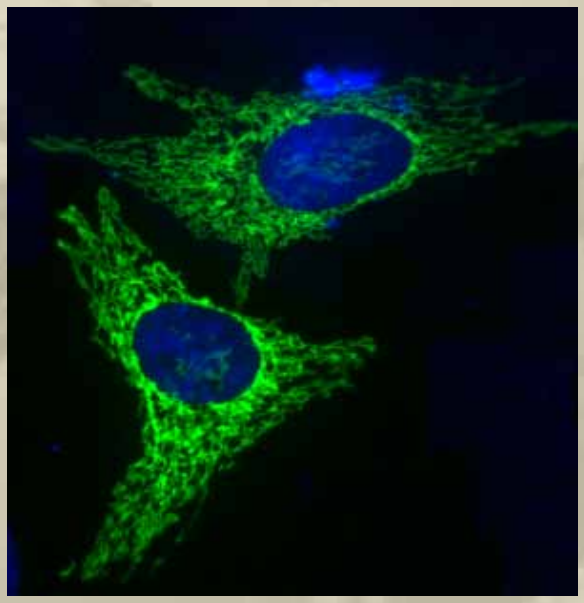
Representative Images

Actin

Peroxisomes

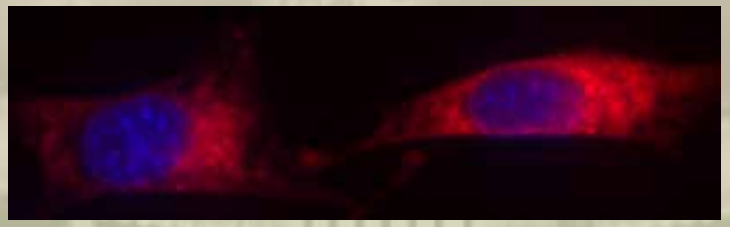


Mitochondria

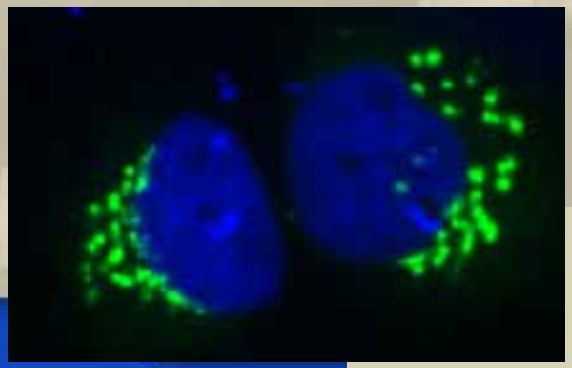


Nucleus

Endoplasmic reticulum



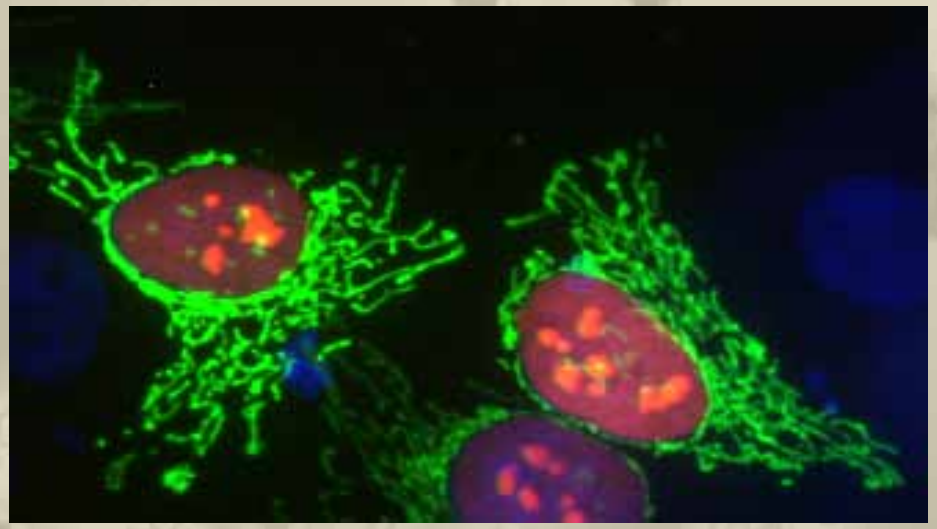
Golgi



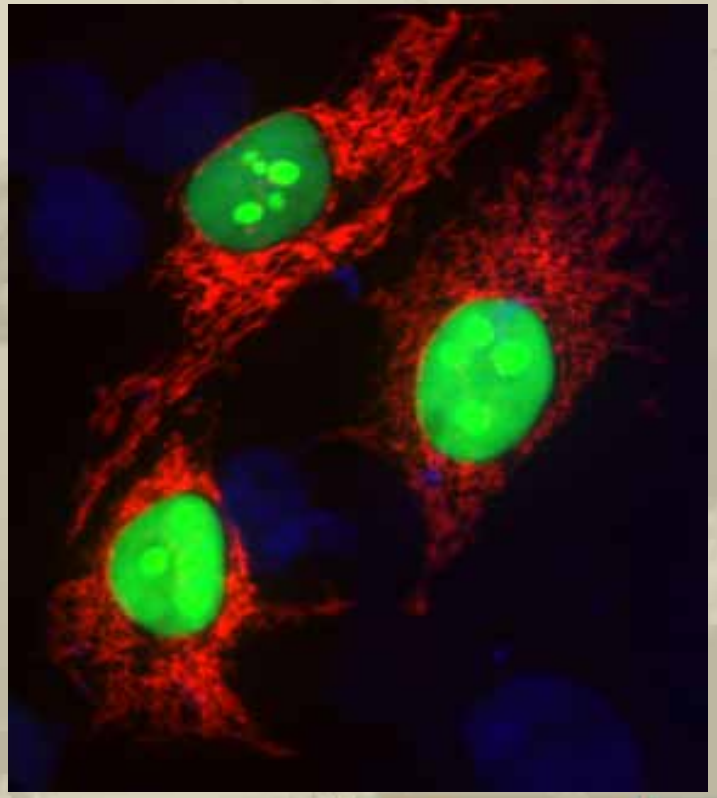
Transfected using FuGENE® 6 Transfection Reagent

Dual FP Transfectants

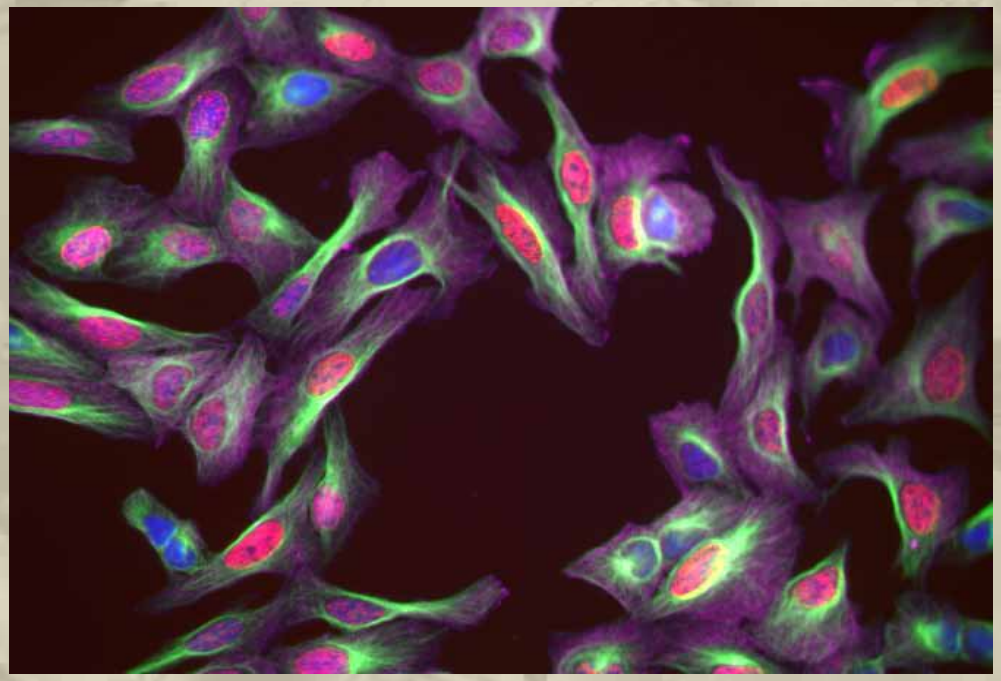
Red nucleus – green mitochondria



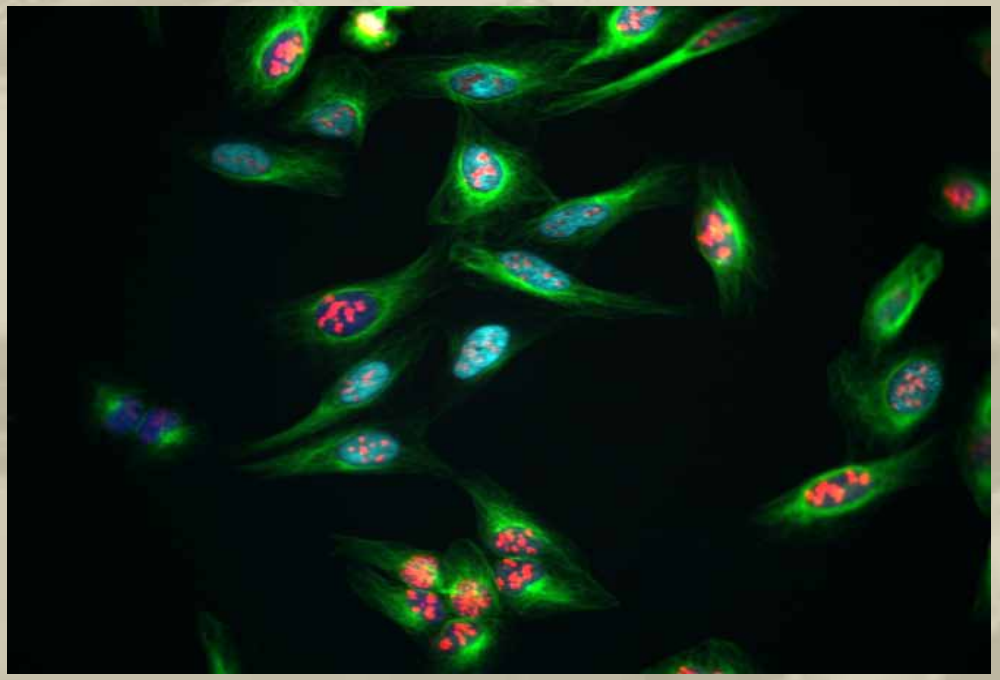
Green nucleus – red mitochondria



Multiplexing with Direct Conjugates



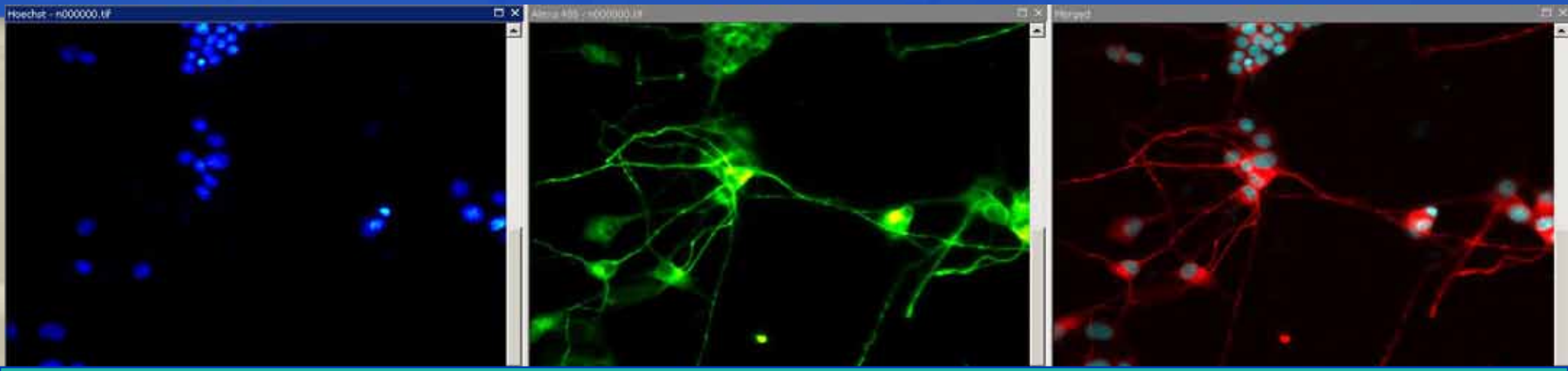
BrdU Alexa 488 (Red)
Actin Alexa 555 (Purple)
β-tubulin Alexa 647 (Green)
Hoechst (Blue)



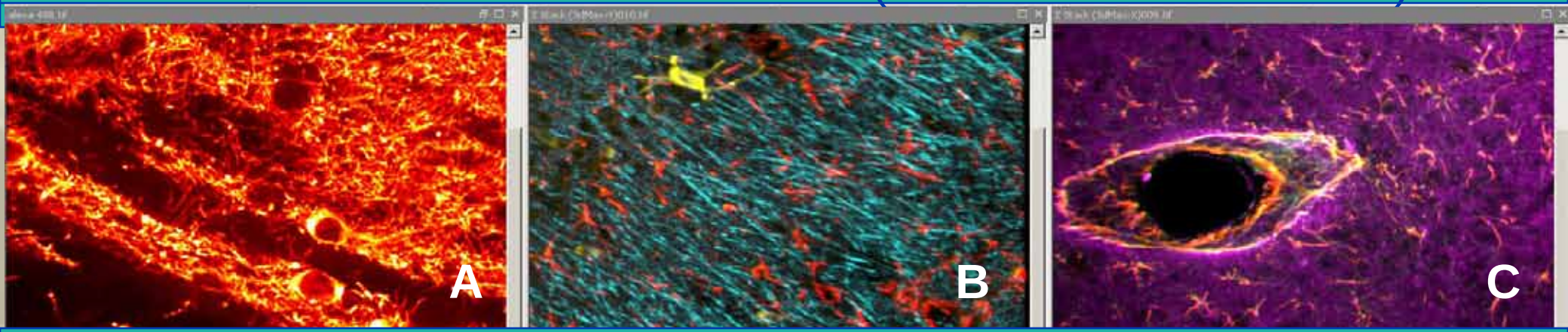
BrdU Alexa 488 (Cyan)
Ki-67 Alexa 555 (Red)
β-tubulin Alexa 647 (Green)
Hoechst (Blue)

Selected Cell Cycle and Cell Morphology Markers

BD Pathway 435 and 855:
Examples for Applications high resolution confocal imaging

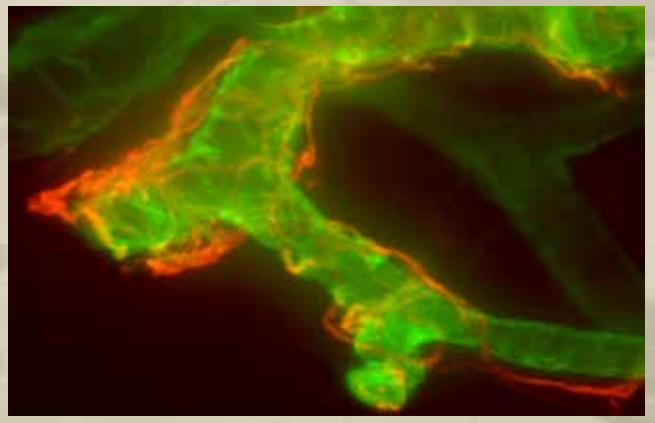
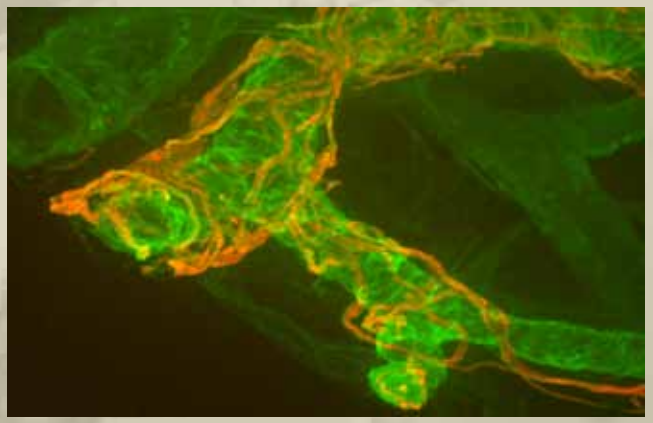
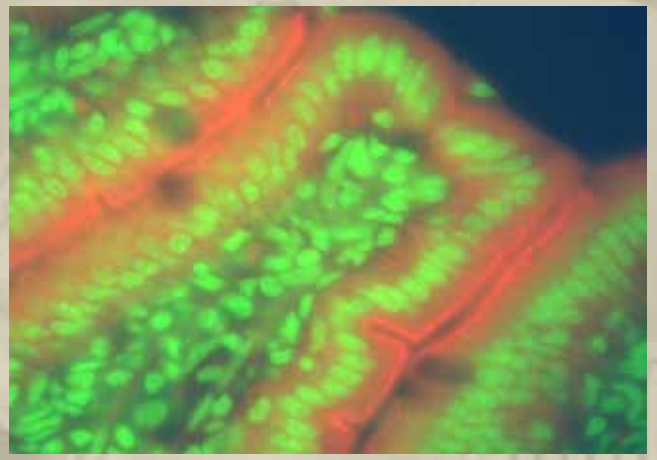
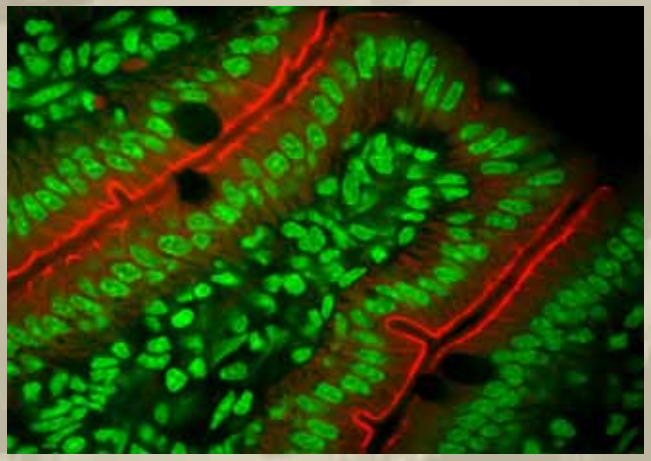


P12 cells diffferentiated with NGF (Hoechst + beta Tubulin)



Rat brain tissue stained with MAP2 (A), GFAP+MAP2+nNOS (B and C)

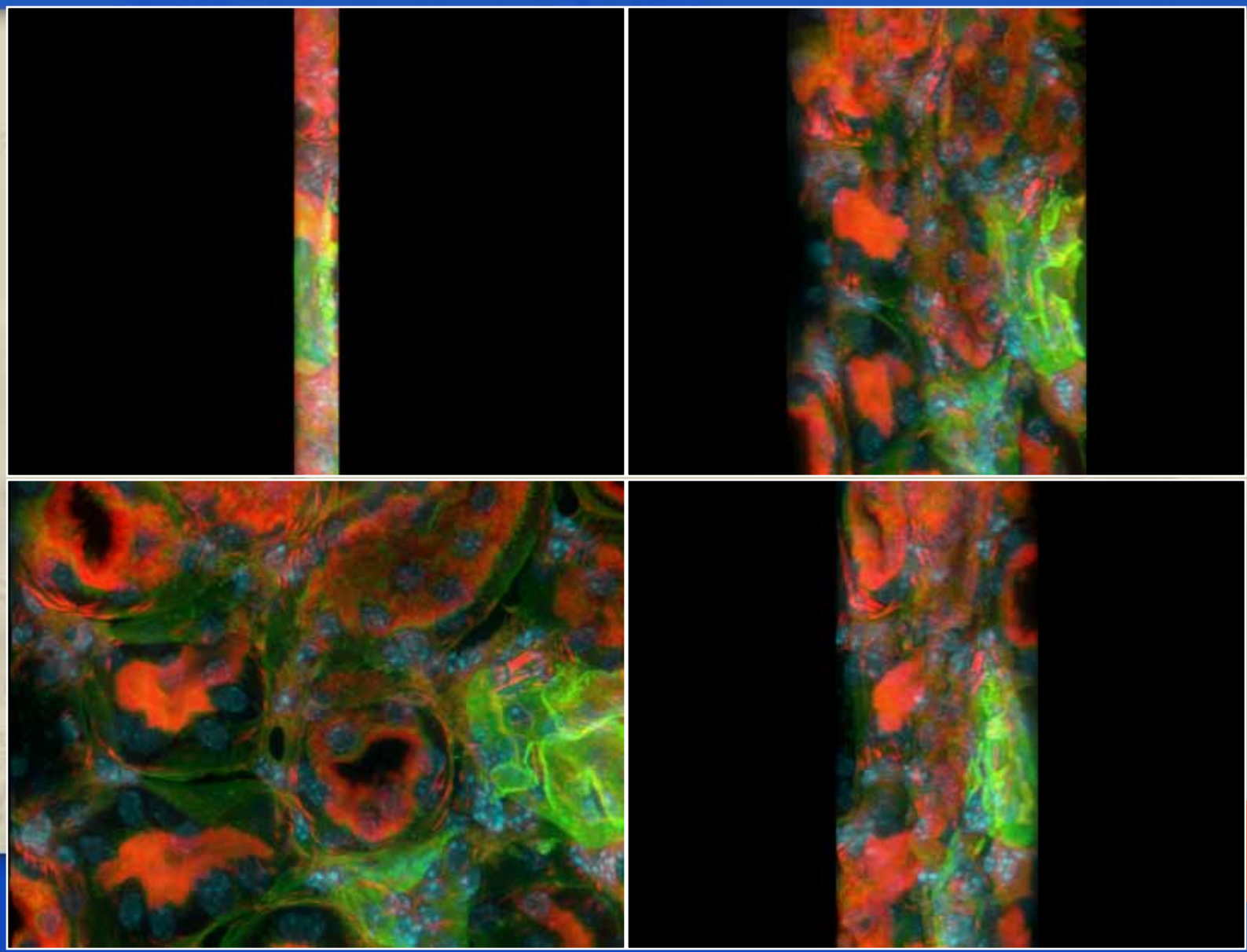
Examples for Applications
Imaging of slides: Comparison wide field versus confocal imaging

	Non Confocal	Confocal
Skin sample Cy3-pan-Neuronal, Cy2-basement membrane marker IV collagen		
Mouse intestine Cytox green Nucleus/ Alexa568-phalloidin		

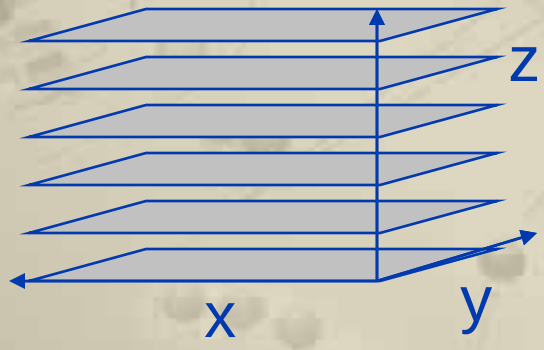
BD Attovision: 3D reconstruction of 60x magnification of a stained mouse kidney section

36 z-sections were taken in the confocal mode with a camera binning of 1 and reconstructed in Attovision

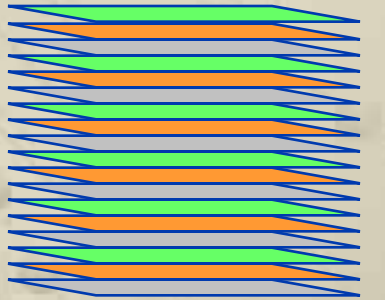
Alexa 488 WGA
Alexa 568 phalloidin
DAPI



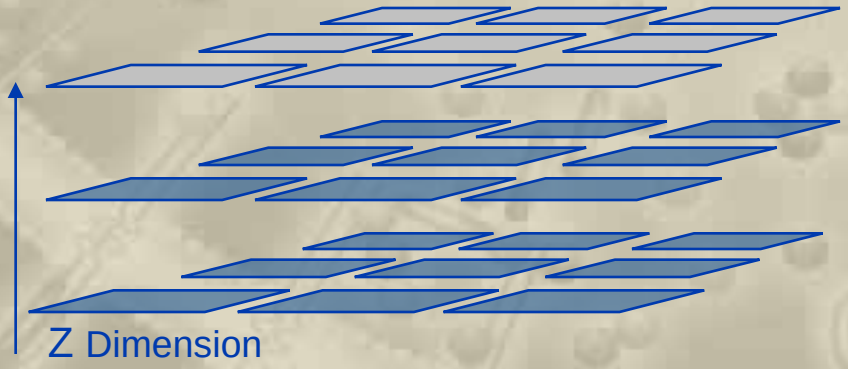
Multidimentional Confocal Imaging Modes: x, y, z, multiple view fields, time, color



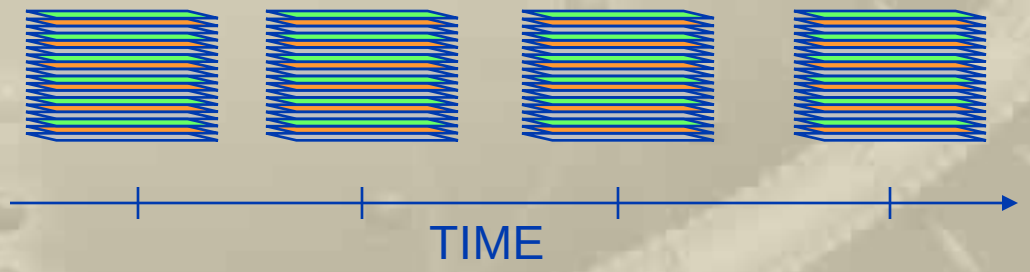
Multiple planes



Multiple dyes: no restriction in number of channels



Multiple view fields: each Z-plane can be montaged



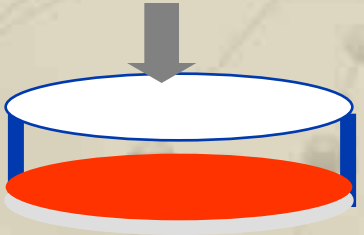
Multiple time points: capture over time in kinetic mode

BD Pathway: Examples for Applications
Angiogenesis : Assay Flow Chart

Day 1

Frozen 96-well Tube Formation product

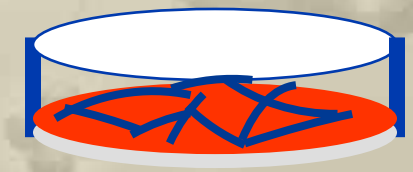
Thaw, 6-24 hours
Gel, 30 min.



Trypsinize & resuspend EC
in assay medium

37°C/5% CO_2 incubation

Day 2



BD Matrigel™ Matrix

Endothelial cells

Angiogenesis
stimulating factors

Fluorescent dye labeling (calcein)

Automated Image Acquisition
& Data Processing

BD

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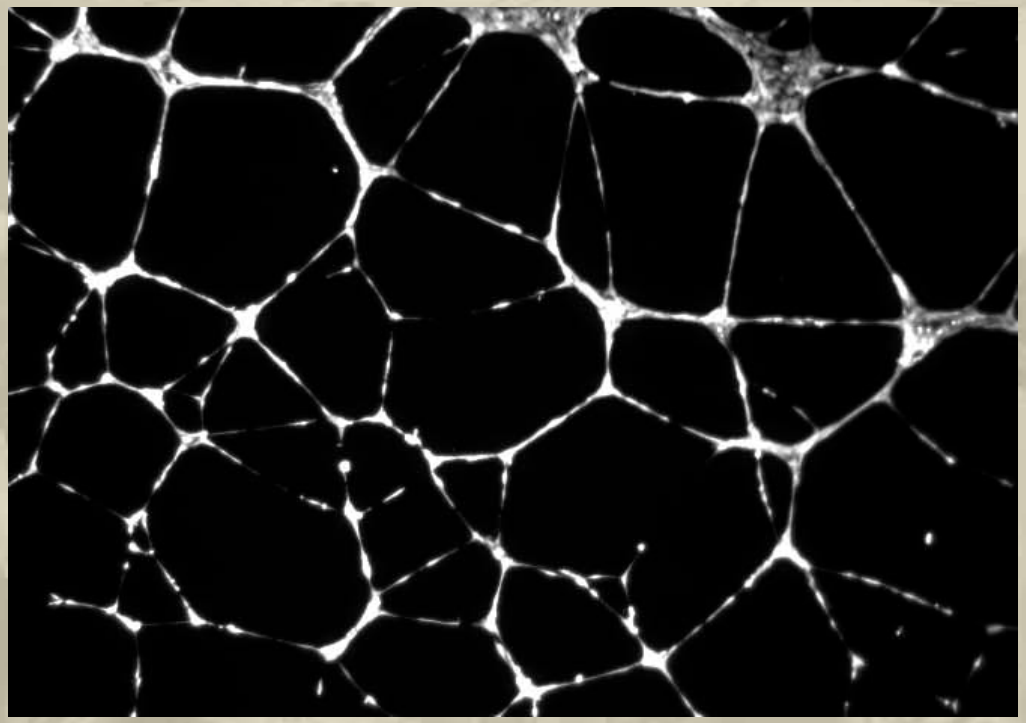
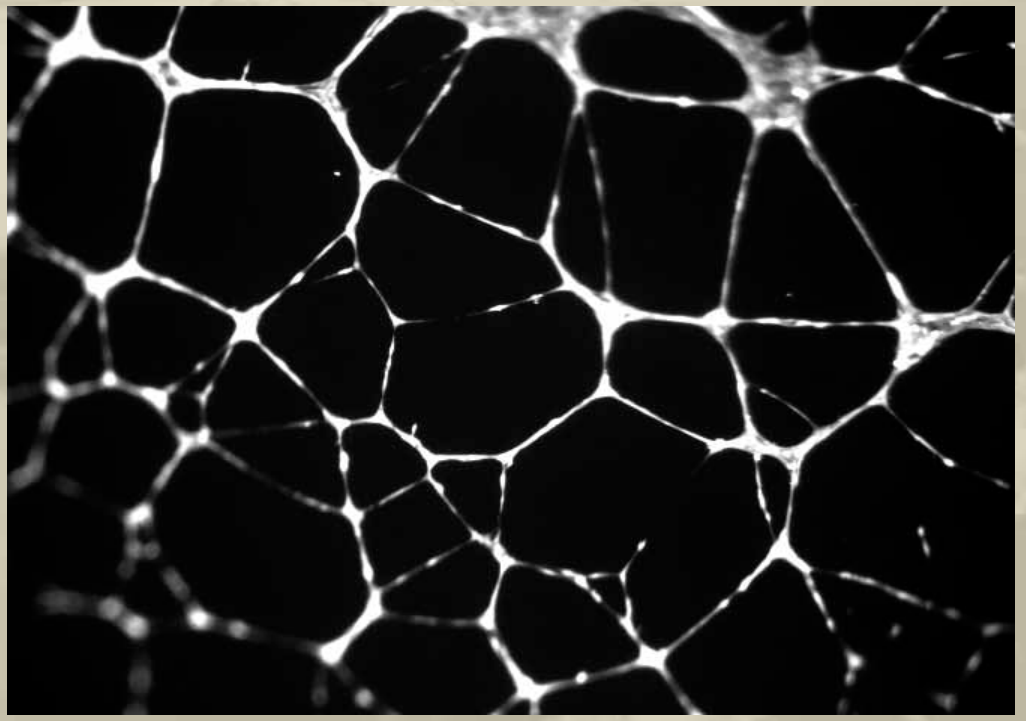
BD Pathway 855: Examples for Applications
Angiogenesis
Confocal imaging mode is essential to analyze 3-dimentional cultures

Confocal



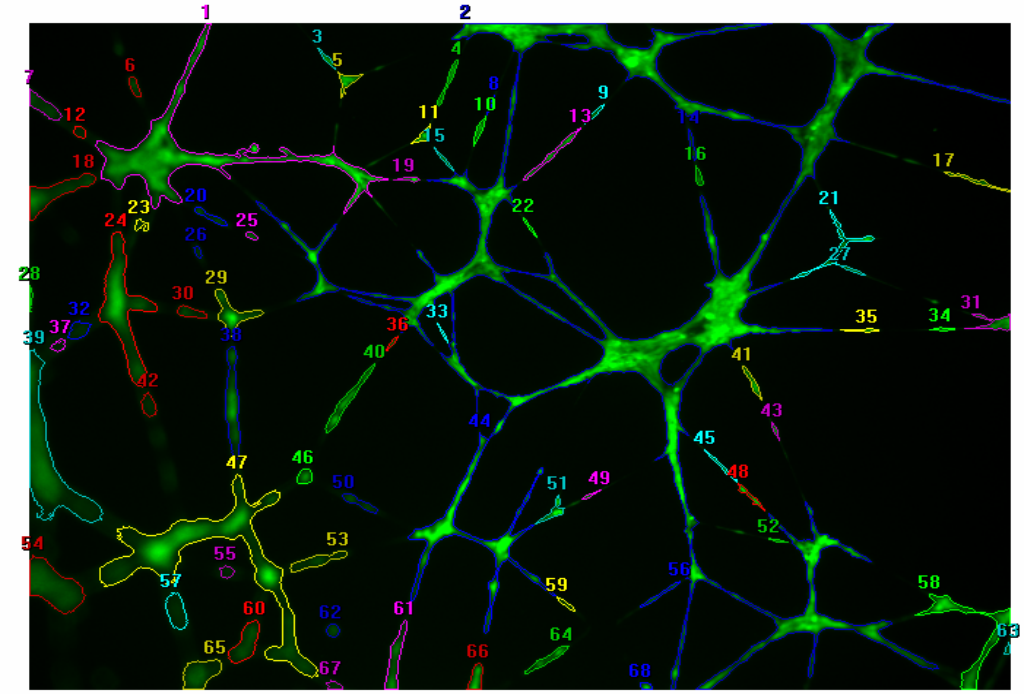
Non-confocal



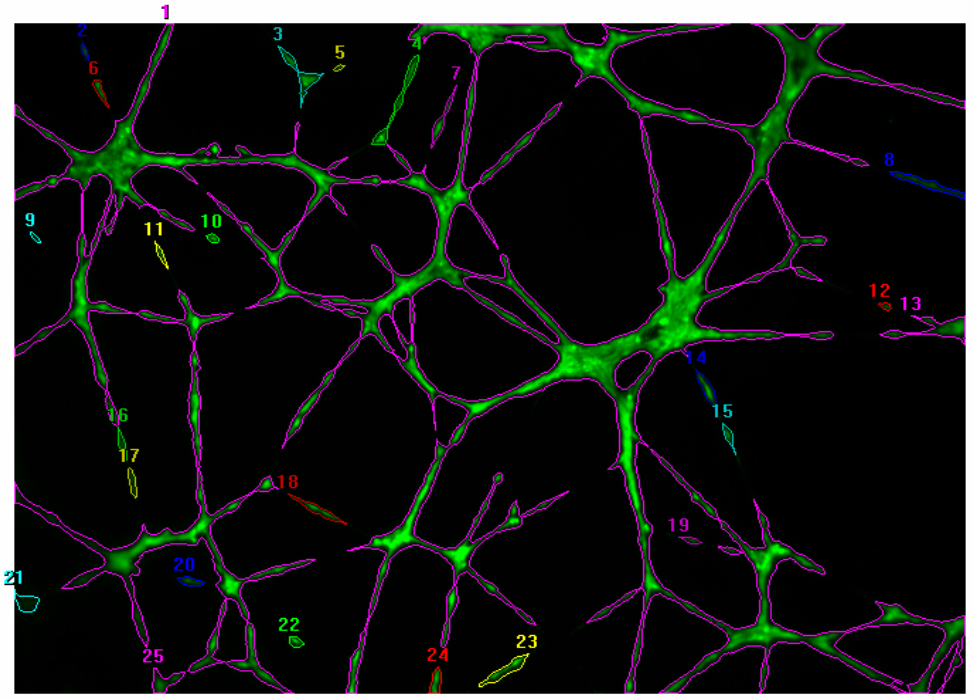
HUVEC-2 cells, stained with Calcien AM,
4X Confocal images show entire network

BD Pathway 855: Examples for Applications
Angiogenesis:
Scientific result without confocality does not reflect situation in culture

Confocal



Non-confocal



Segmentation result:

Discrete network islets

Complete network

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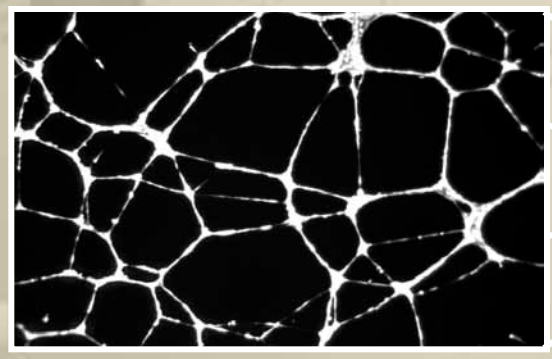
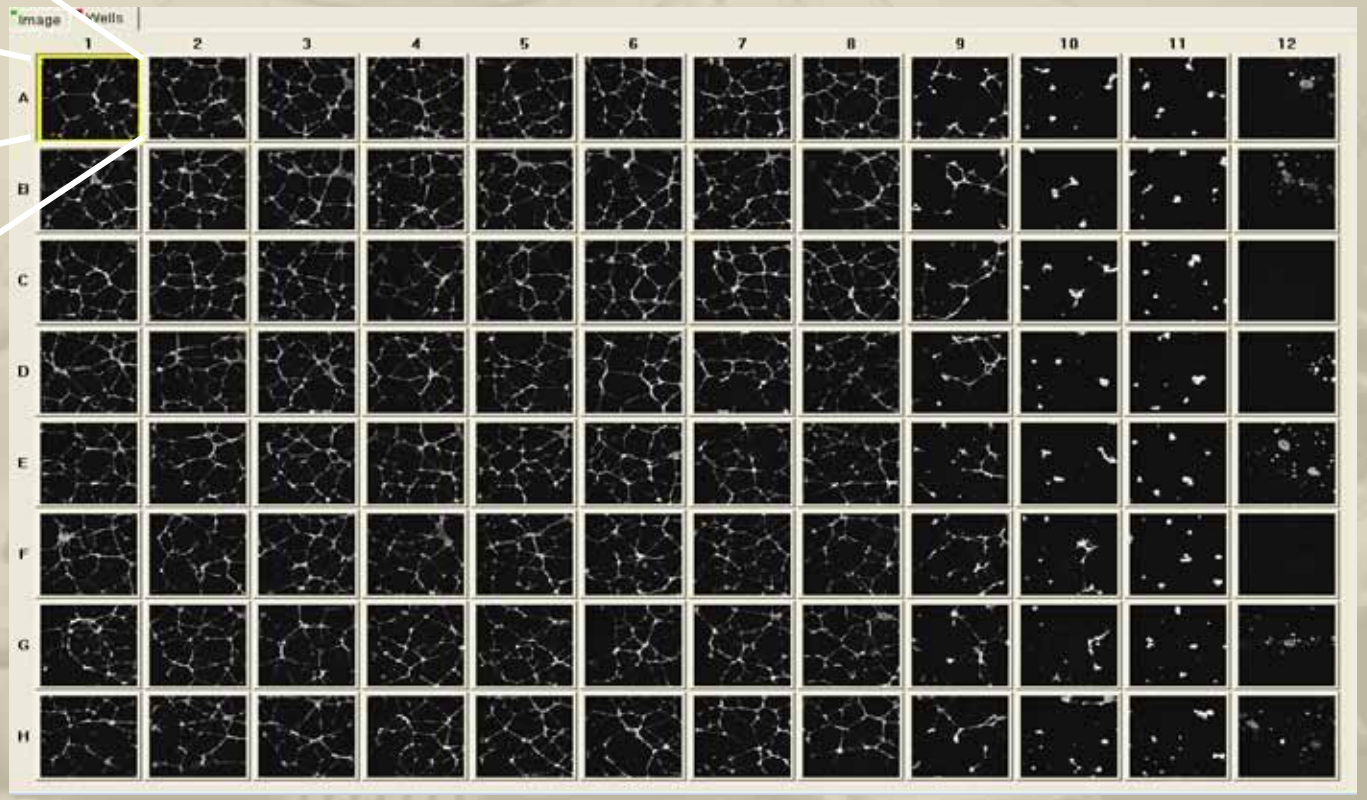
BD Pathway 855: Examples for Applications
Angiogenesis - Dose Response to Suramin





0 → 160 uM

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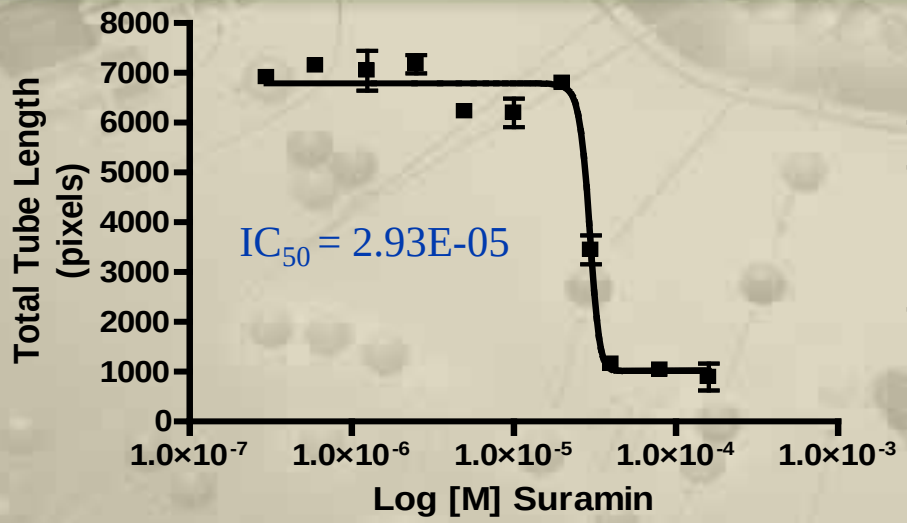
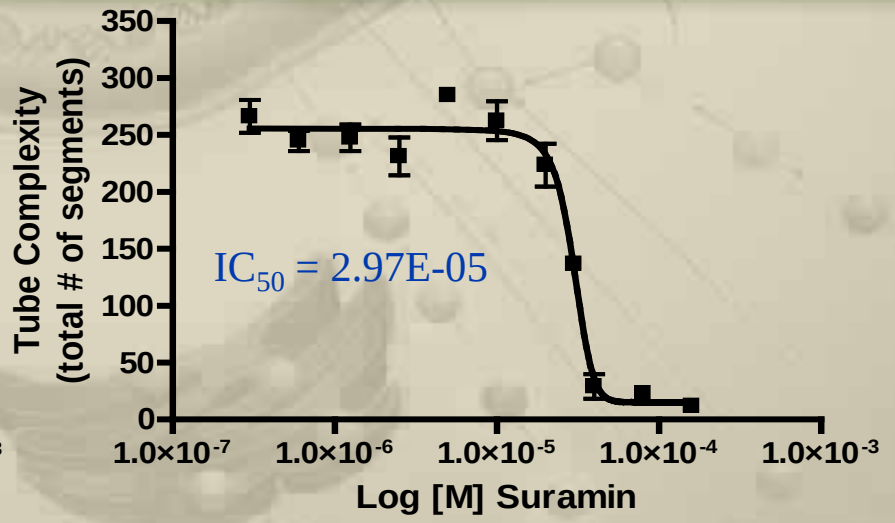
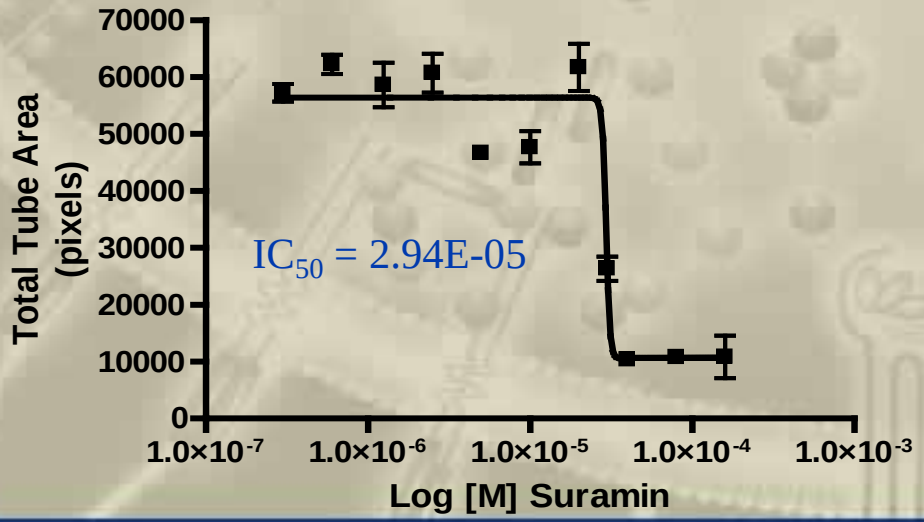
BD Pathway 855: Examples for Applications
Angiogenesis: Different Analysis Parameters - Similar Results







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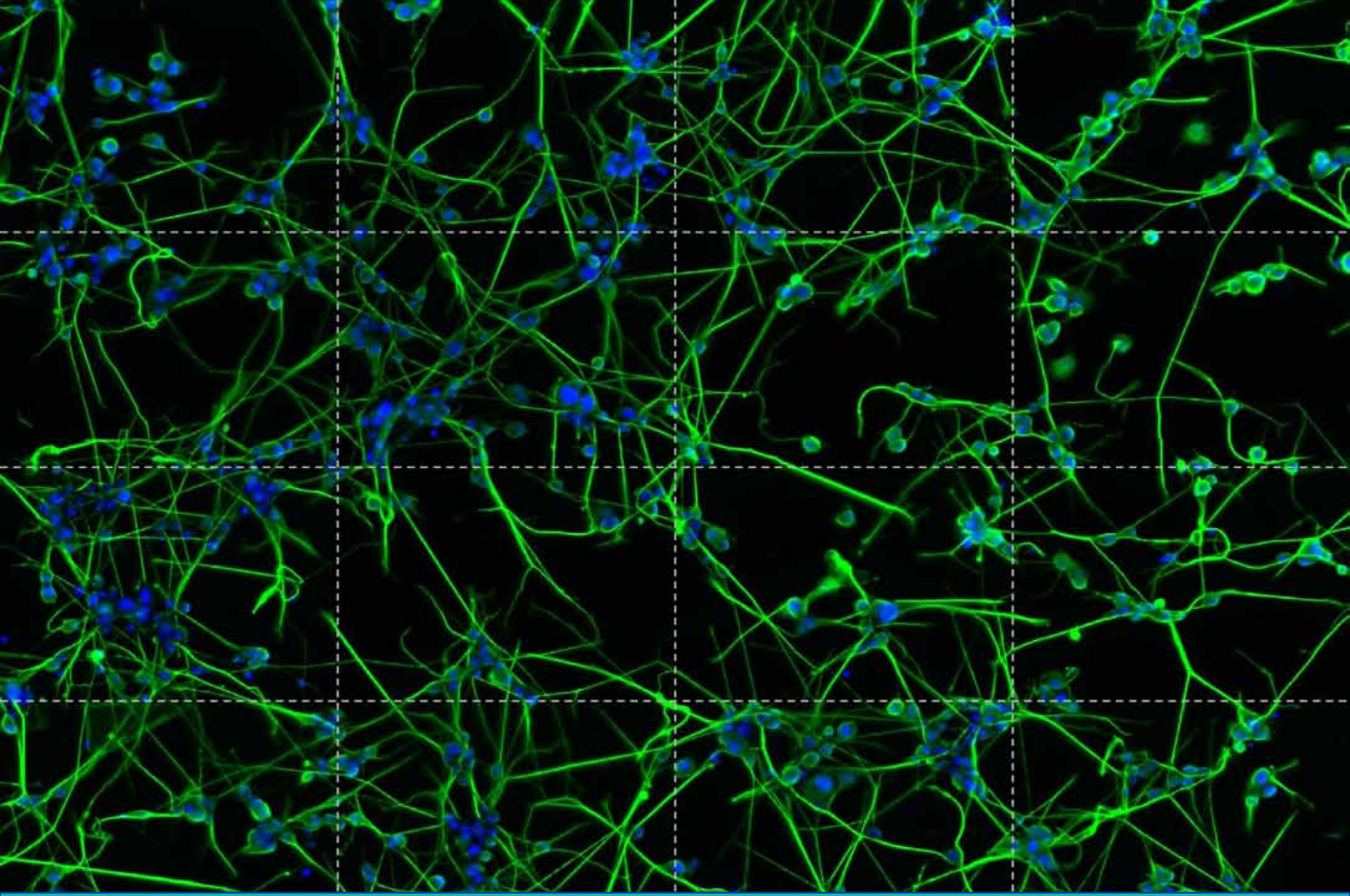
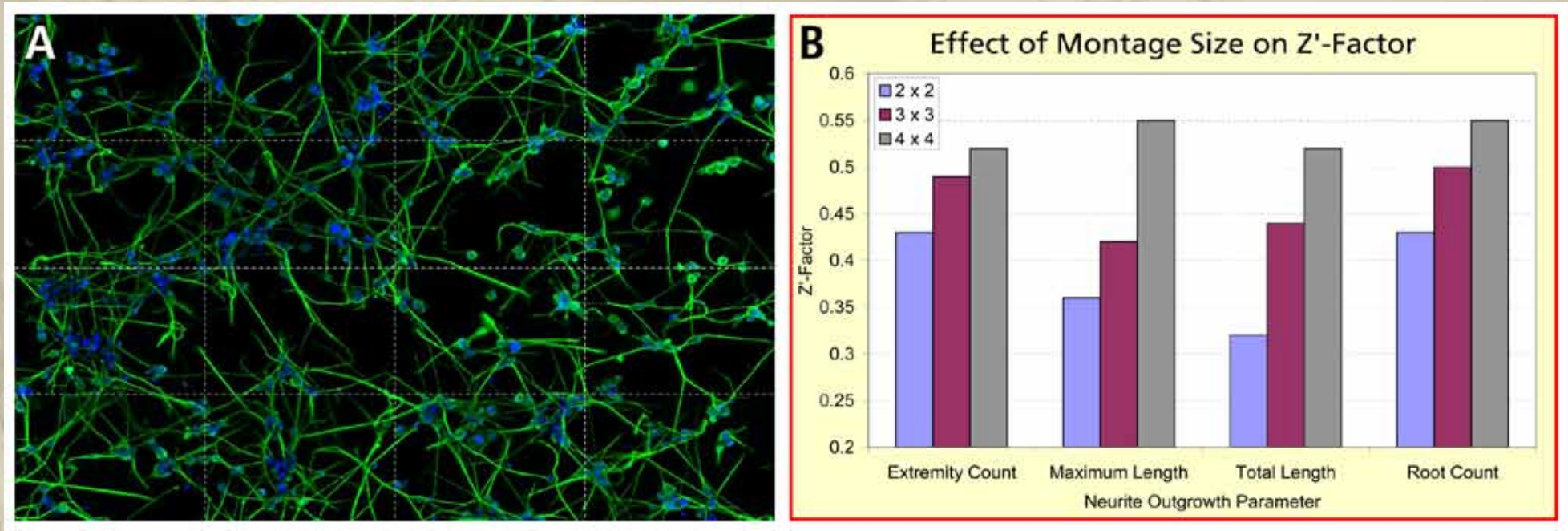


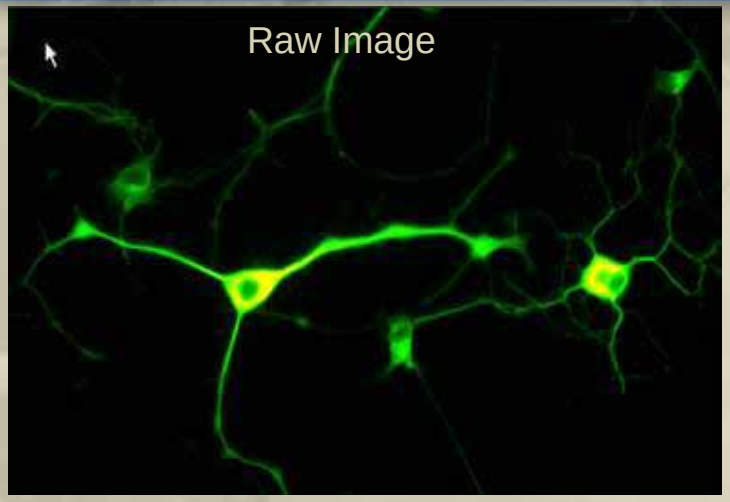
P12 cells diffferentiated with NGF (Hoechst + beta Tubulin, 20x, collapsed stack, 4x4 montage)

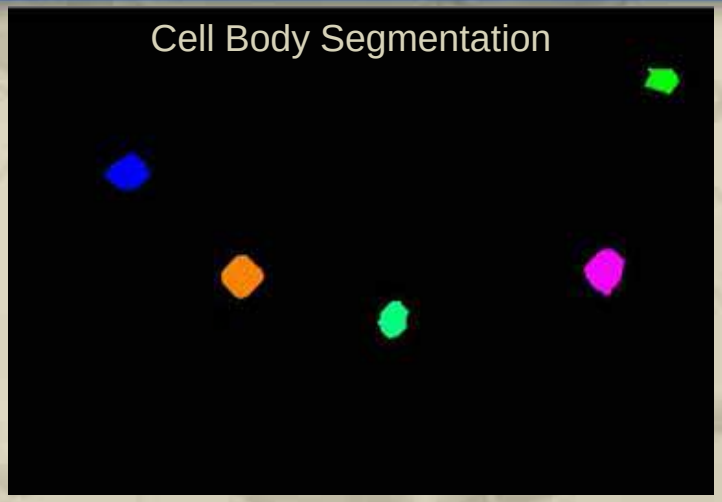
Effect of Montage on Z′-Factor

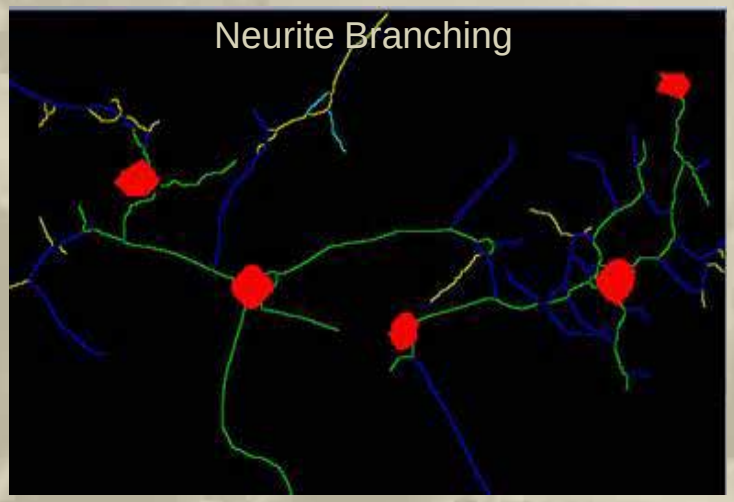


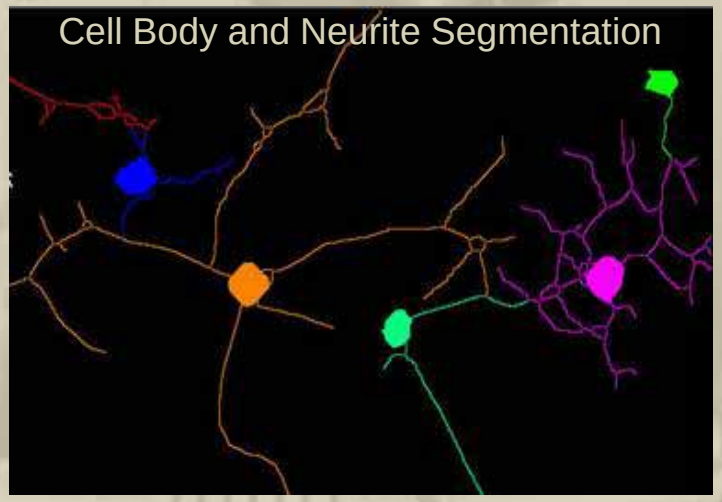
As a representative assay, data from a Neurite Outgrowth assay is shown.
Increasing the montage size improved the Z'-factor.

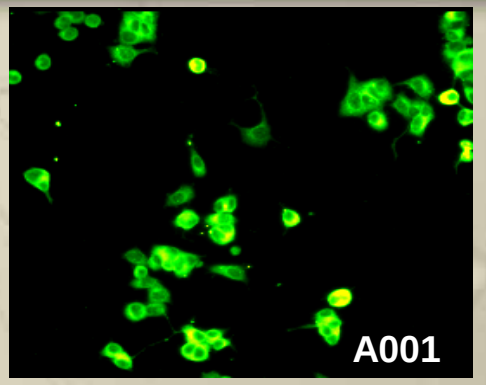
Example: Measurement of Morphology to analyze Neurite Outgrowth

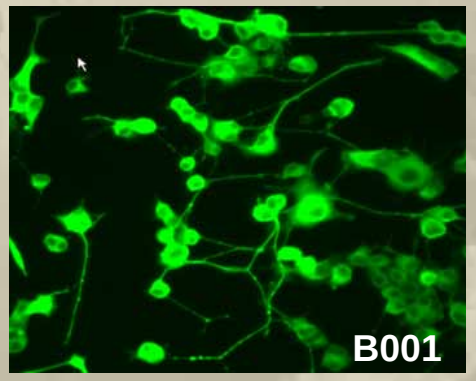


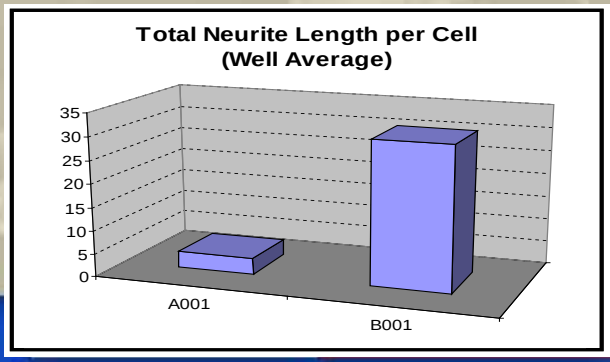






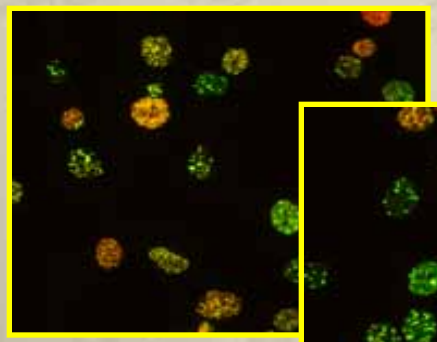




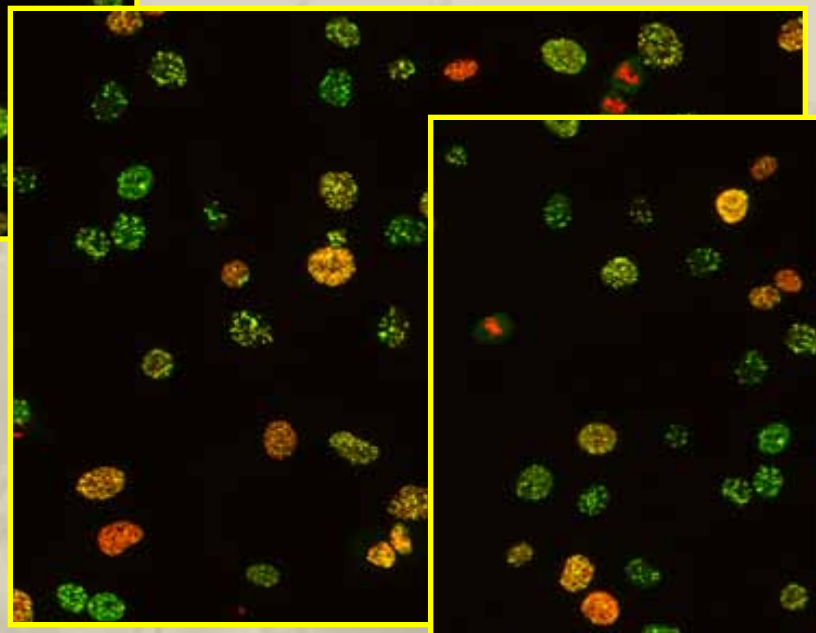


Examples for Applications
DNA strand break analysis: High-Precision Montage – no stitching necessary

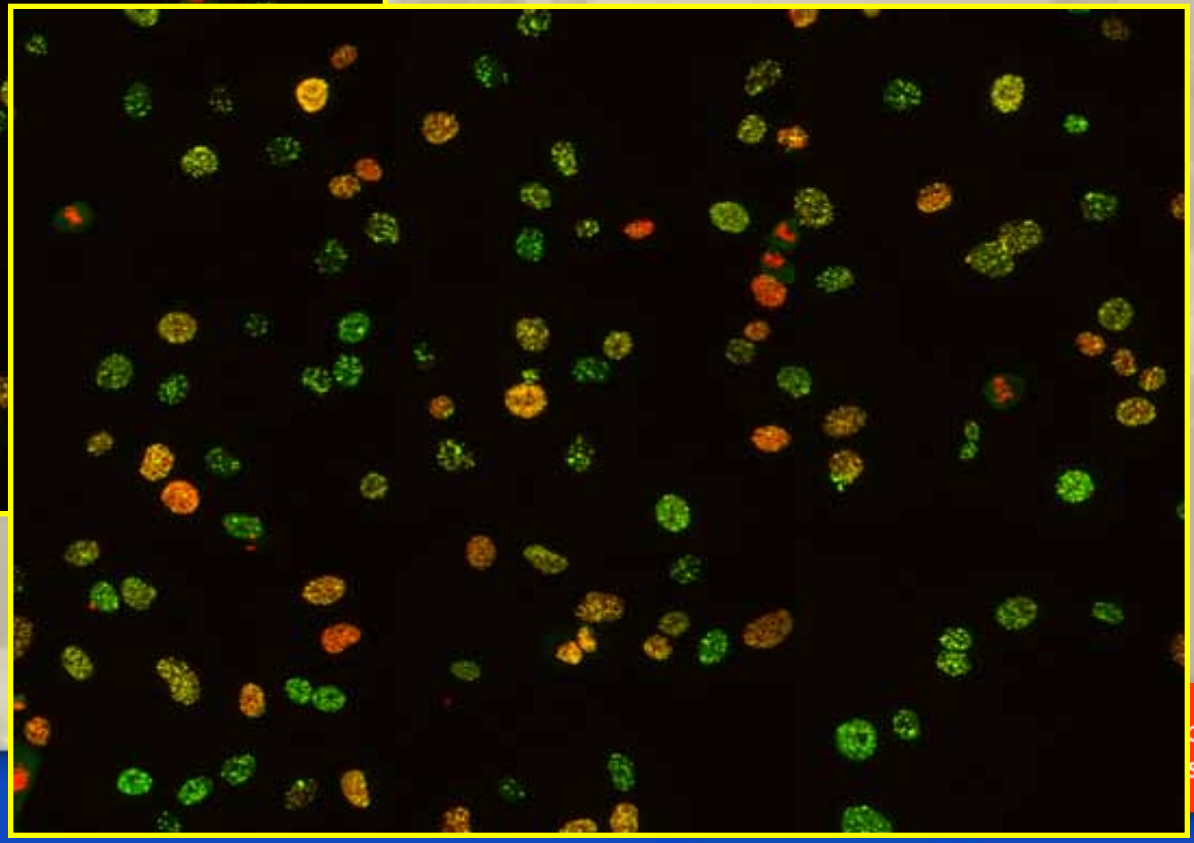
Single image field ~ 20 cells



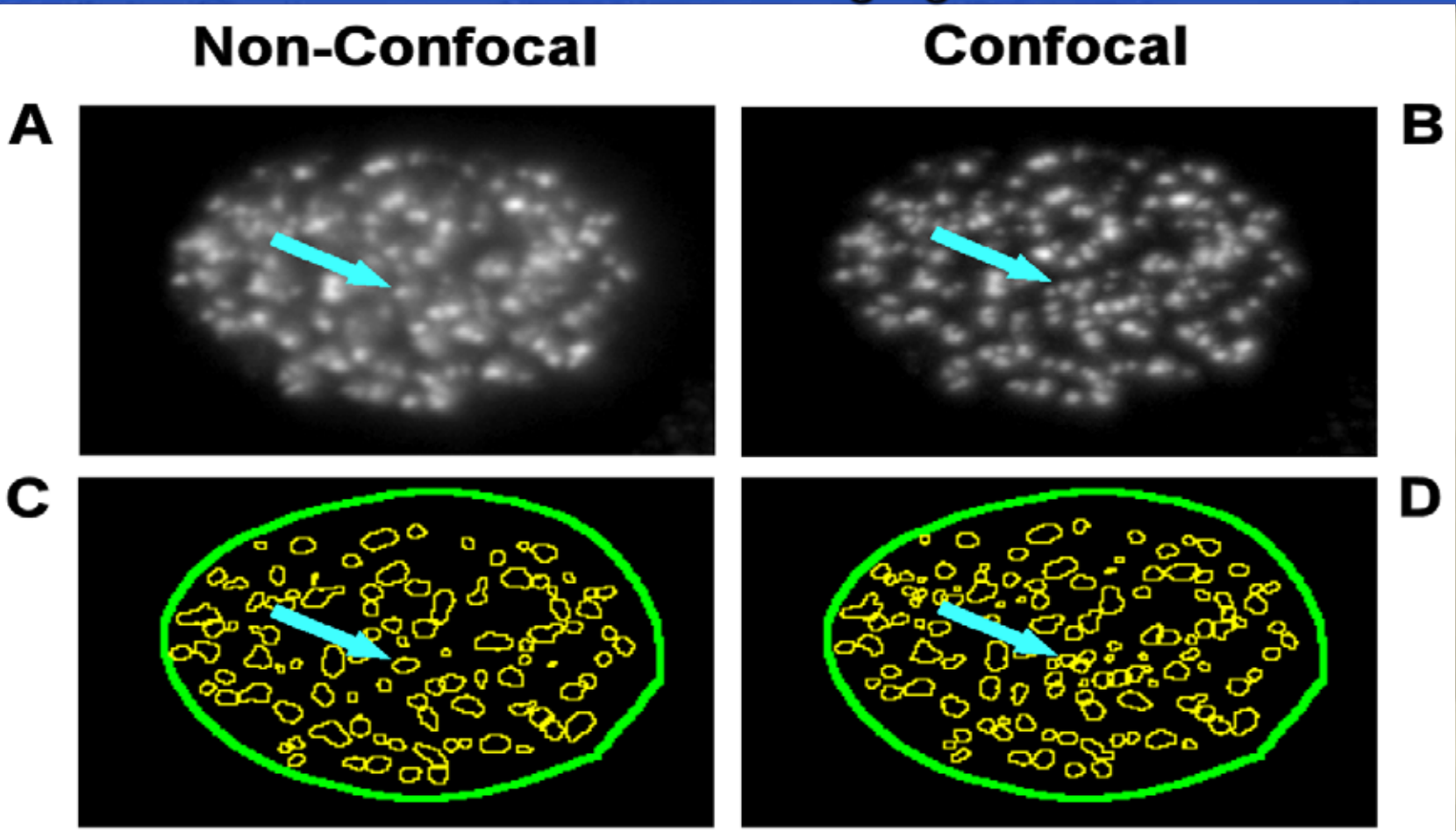
2x2 Montage ~ 80 cells



3x3 Montage ~ 180 cells



BD Pathway: Examples for Applications
DNA strand break analysis*. Comparison wide field versus confocal imaging

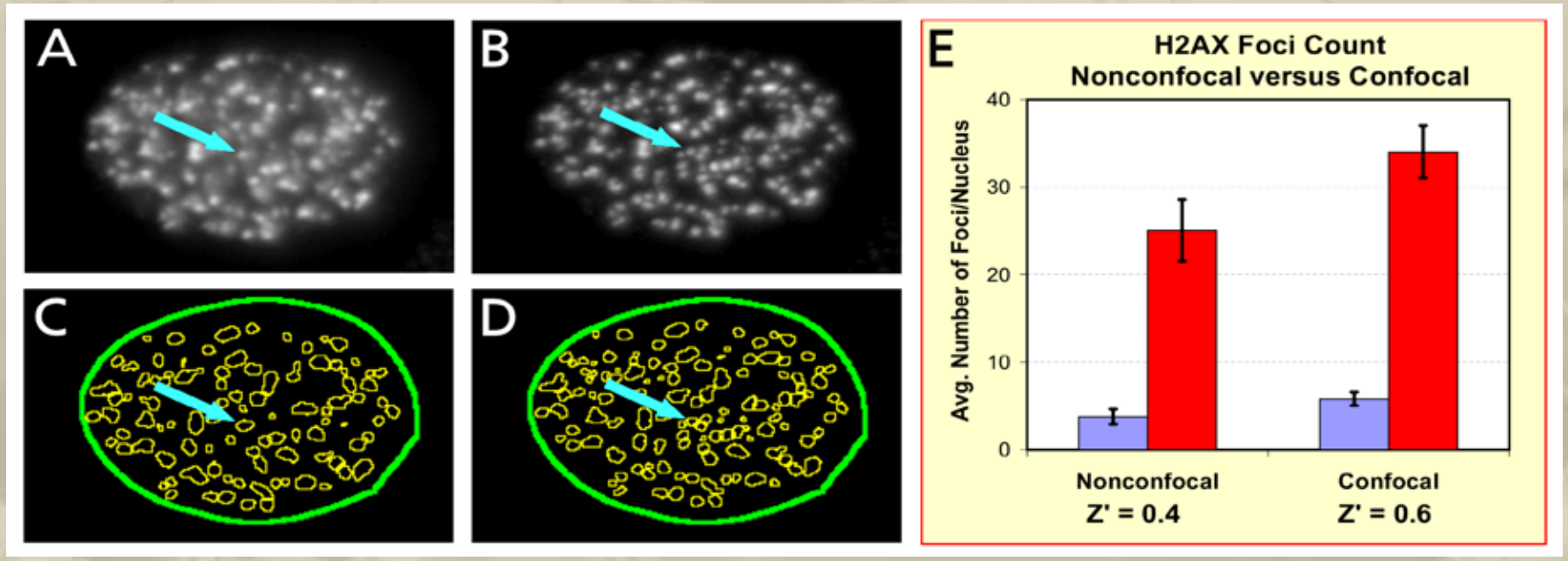


DNA damage Antibody (Anti H2AX)

Foci detection

40x objective

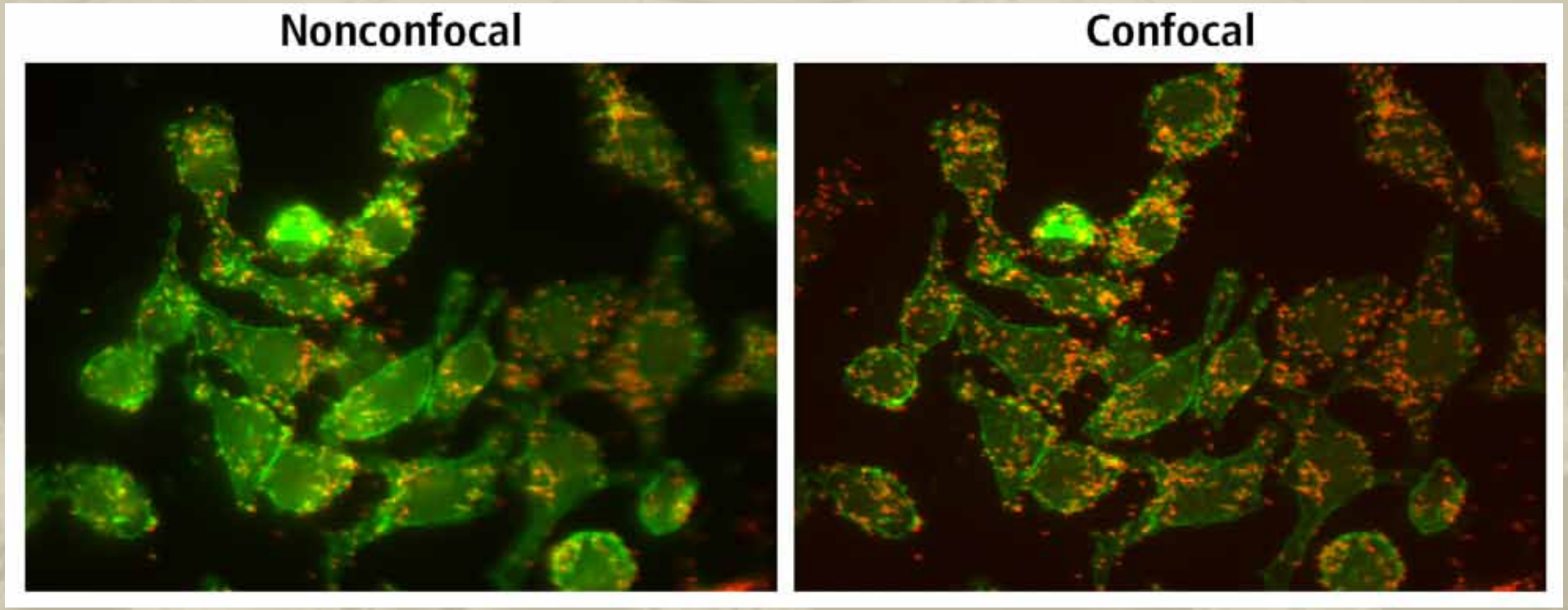
Confocal vs. Nonconfocal Imaging



As a representative assay data from a DNA damage assay is shown.
Confocal imaging improves resolution and data quality.

BD Pathway: Examples for Applications
Bacterial infection: High Resolution for Bacteria counting and colocalisation analysis

Confocal Collapsed Stack Improves Image Quality
Individual Bacteria Can be Quantitated



Comparison of nonconfocal and confocal image acquisition modes. Pseudocolored merged images (40x, 0.9 NA) of macrophages after infection with *L. monocytogenes*. Macrophage actin is green and bacteria are red. Colocalized signals appear as merges of these color channels. Panel A, single plane nonconfocal image. Panel B, confocal collapsed stack of the same image field.

BD Pathway: Examples for Applications
Prerequisite for bacteria counting: precise determination of objects.
Object-within-object detection algorithmus

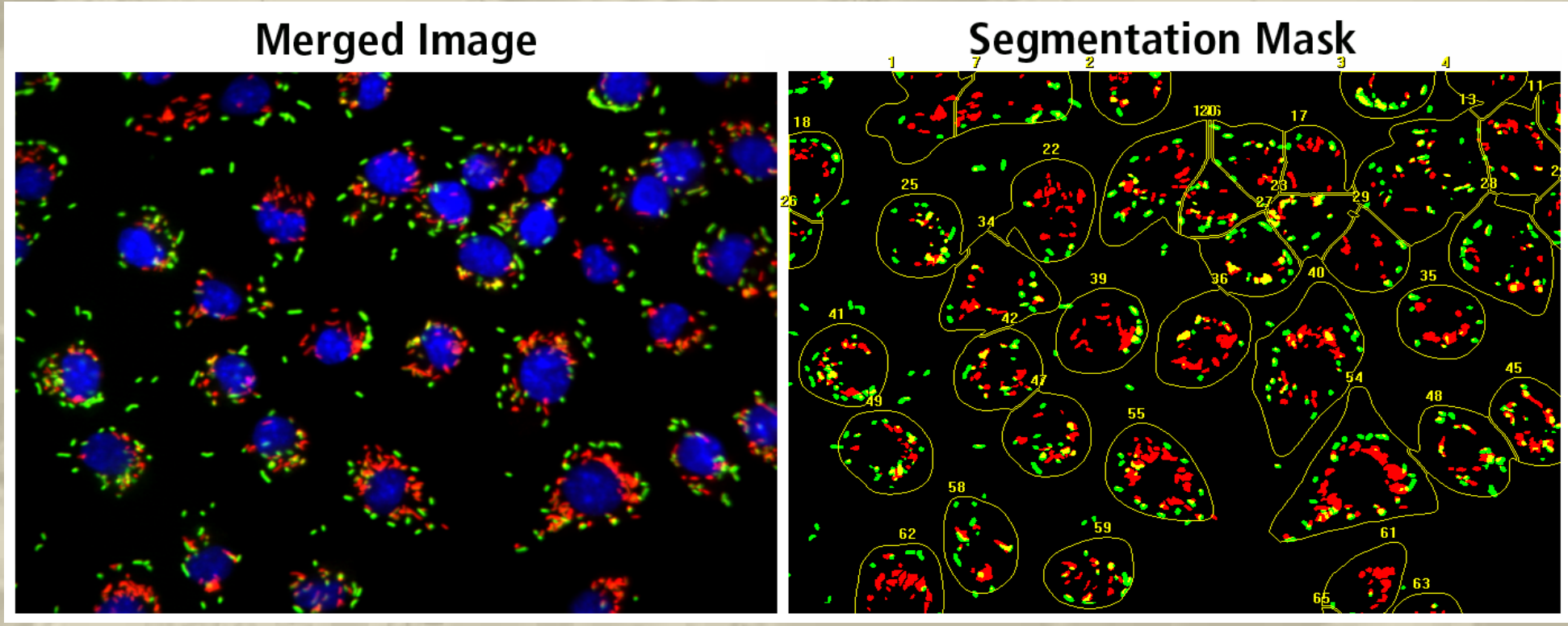


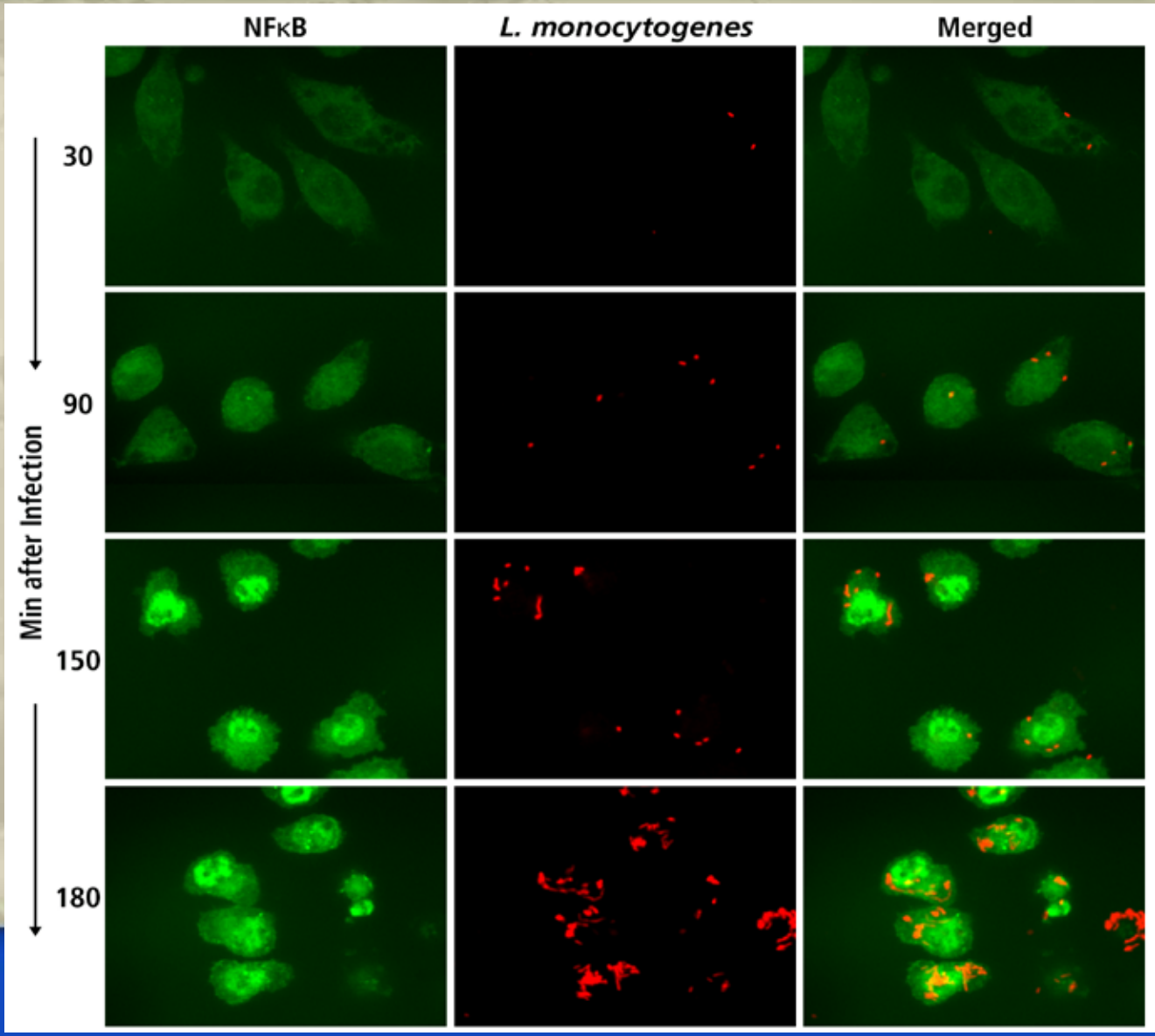
Image mask created after Sub Object counting. Panel A, pseudocolored merged single field confocal collapsed stack image (40x, 0.9 NA) of macrophages after infection with *L. monocytogenes.* Macrophage nuclei are blue, bound bacteria are green, and total bacteria are red (whole-cell stain channel not shown). Colocalized signals appear as merges of these color channels. Panel B, segmentation mask of the same image generated after Sub Object analysis depicting macrophage boundaries (yellow line), bound bacteria (green), internal bacteria (red), colocalized bacteria (yellow), and numbered cellular ROIs.

BD Pathway 855: Examples for Applications Mulitiplexing for High-Content Analysis

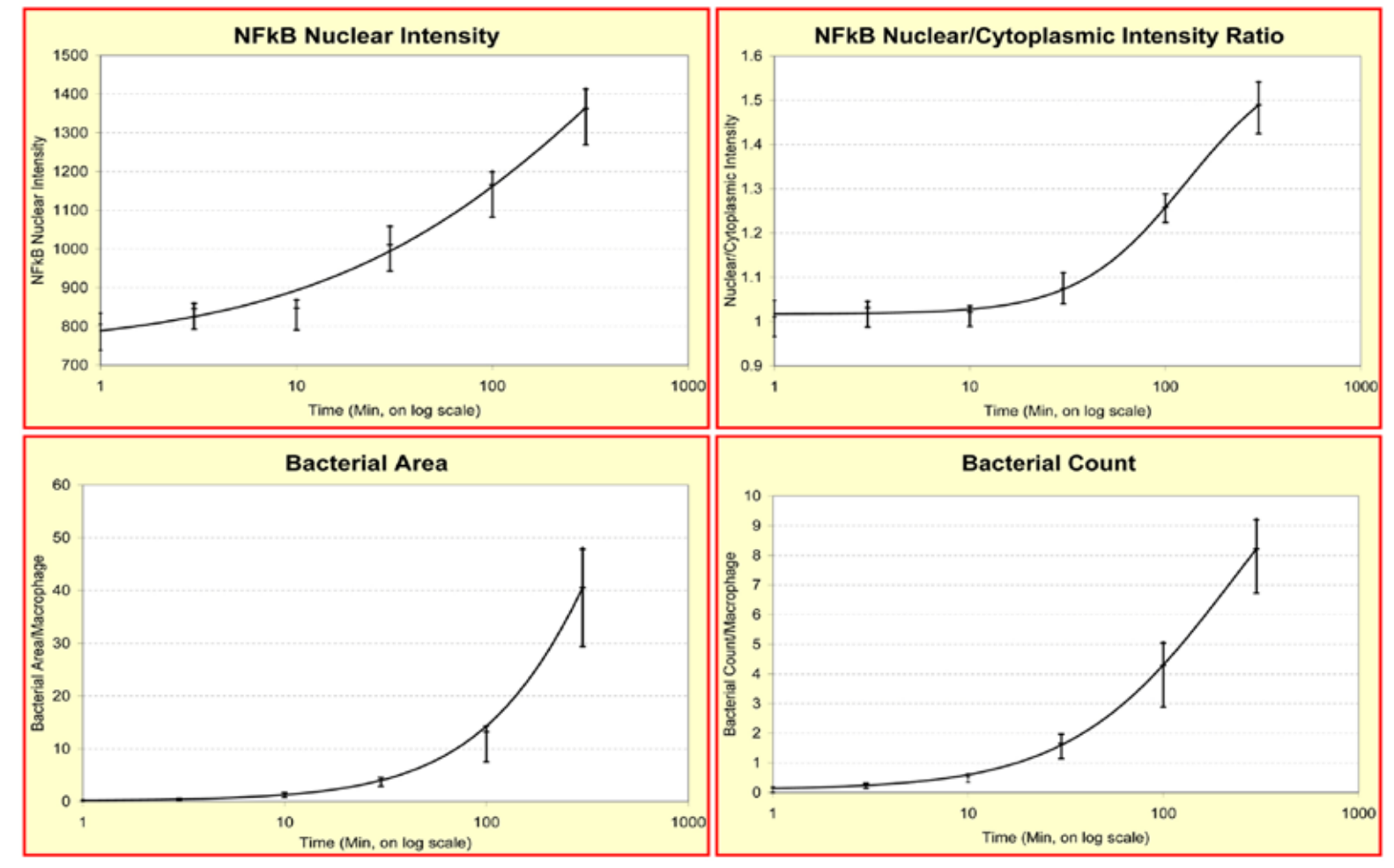
Host Cell-Signaling Activation in Response to Infection

NFkB Translocation

Multiplexing NF-kB and bacterial replication assays. Panel A, representative pseudocolored cropped confocal collapsed stack images (40x, 0.9 NA) at different time points after infection. NF-kB protein is green, bacteria are red, and colocalized signals appear as merges of these color channels. Panels B and C, NF-kB intensity in the nucleus and the ratio of NF-kB nuclear-to-cytoplasmic intensity, respectively (n = 3 wells).



BD Pathway: Examples for Applications
Quantification of Multiplexed Data:
Translocation, bacterial count and area of macrophage covered with bacteria



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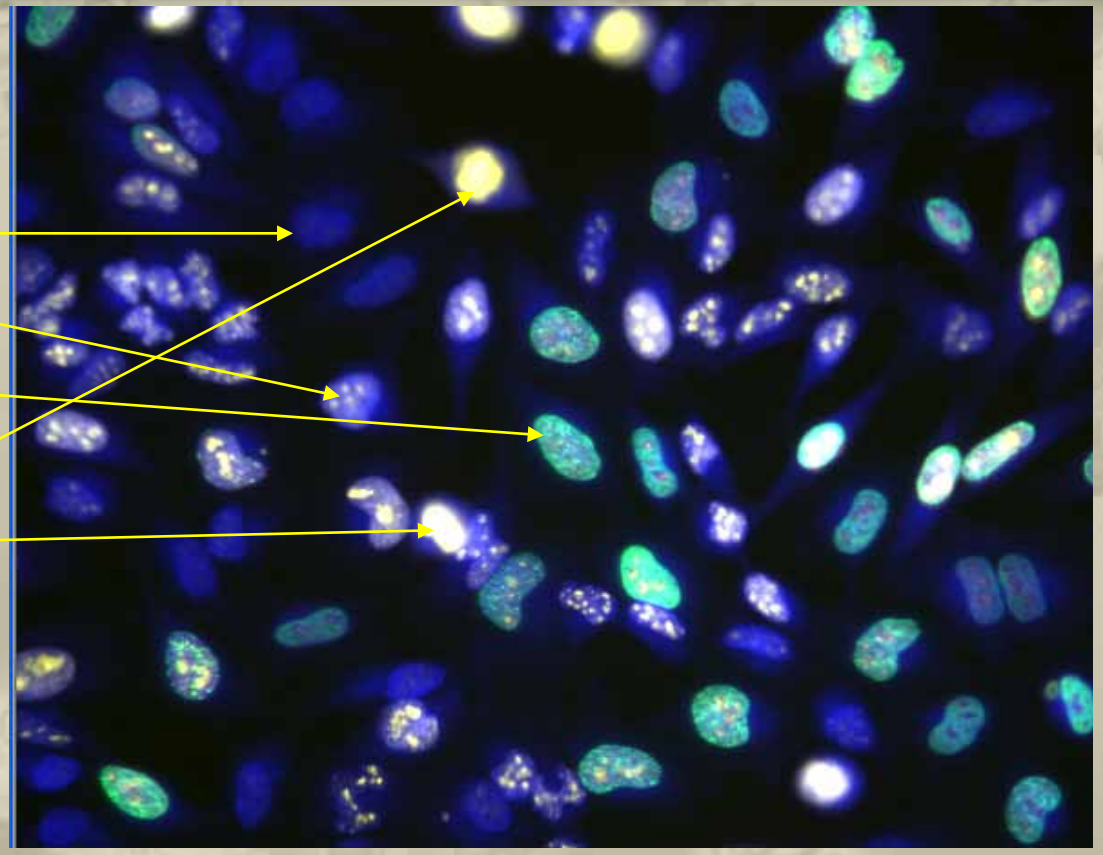
BD Pathway: Examples for Applications : Multiplexing multiple stainings (primary conjugated Antibodies) to enable Cell Cycle Assays

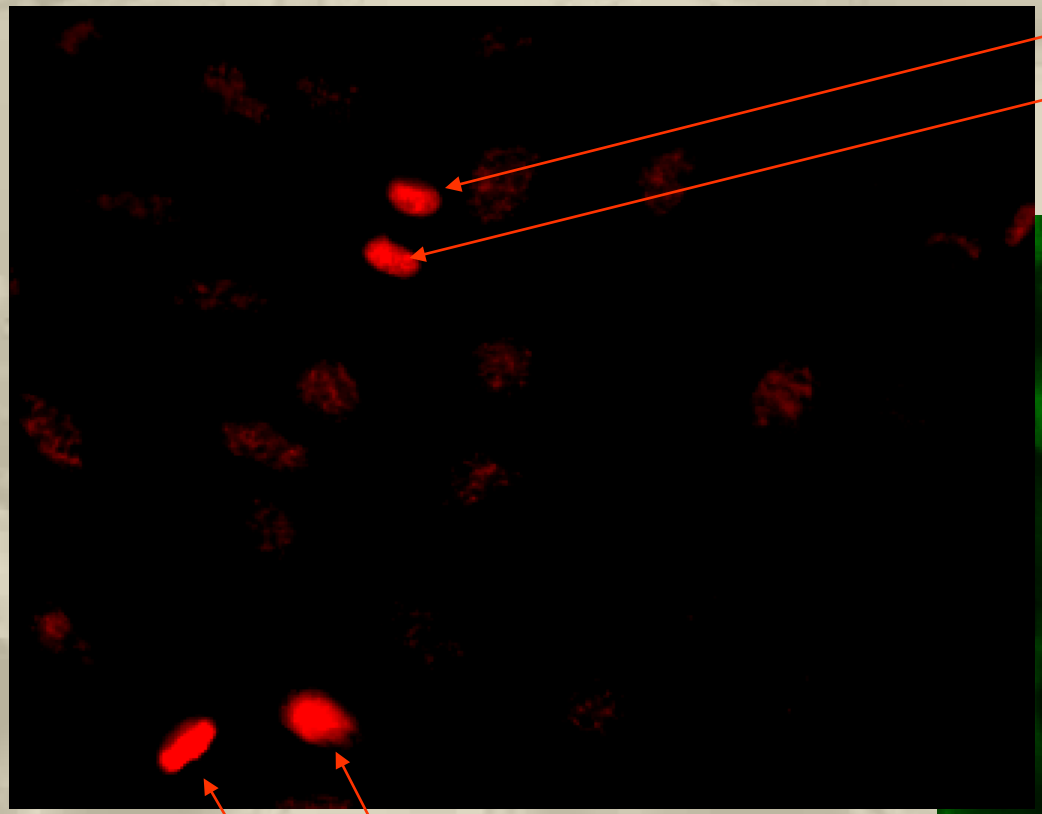


	BrdU	pHis H3	Ki-67
G0	–	–	–
G1	–	–	punctate
S	+	–	punctate
G2	–	–	diffuse
M	–	+	diffuse

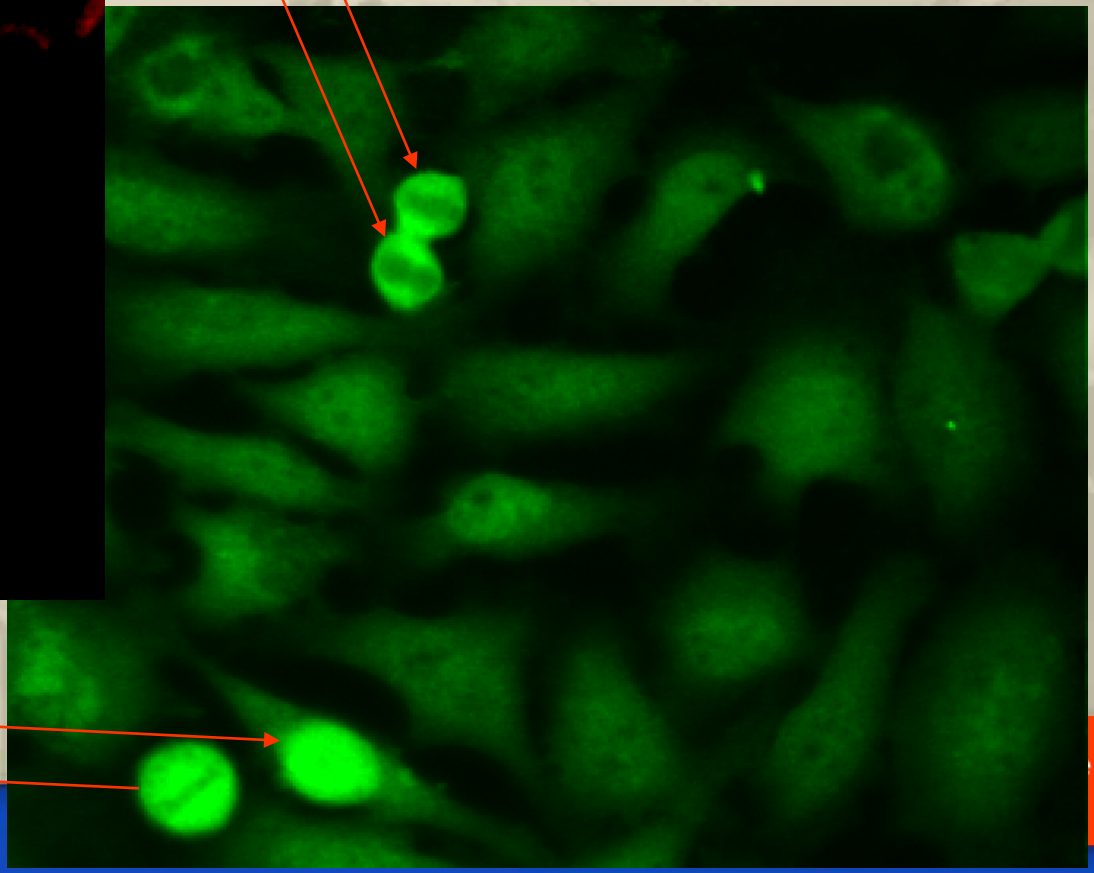
BrdU Alexa 488 (green)
pHistone H3 Alexa 647 (white)
Hoechst (blue)
Ki-67 Alexa 555 (yellow)

How Cell Cycle Can Affect Other Results

Phospho Histone H3 Marker

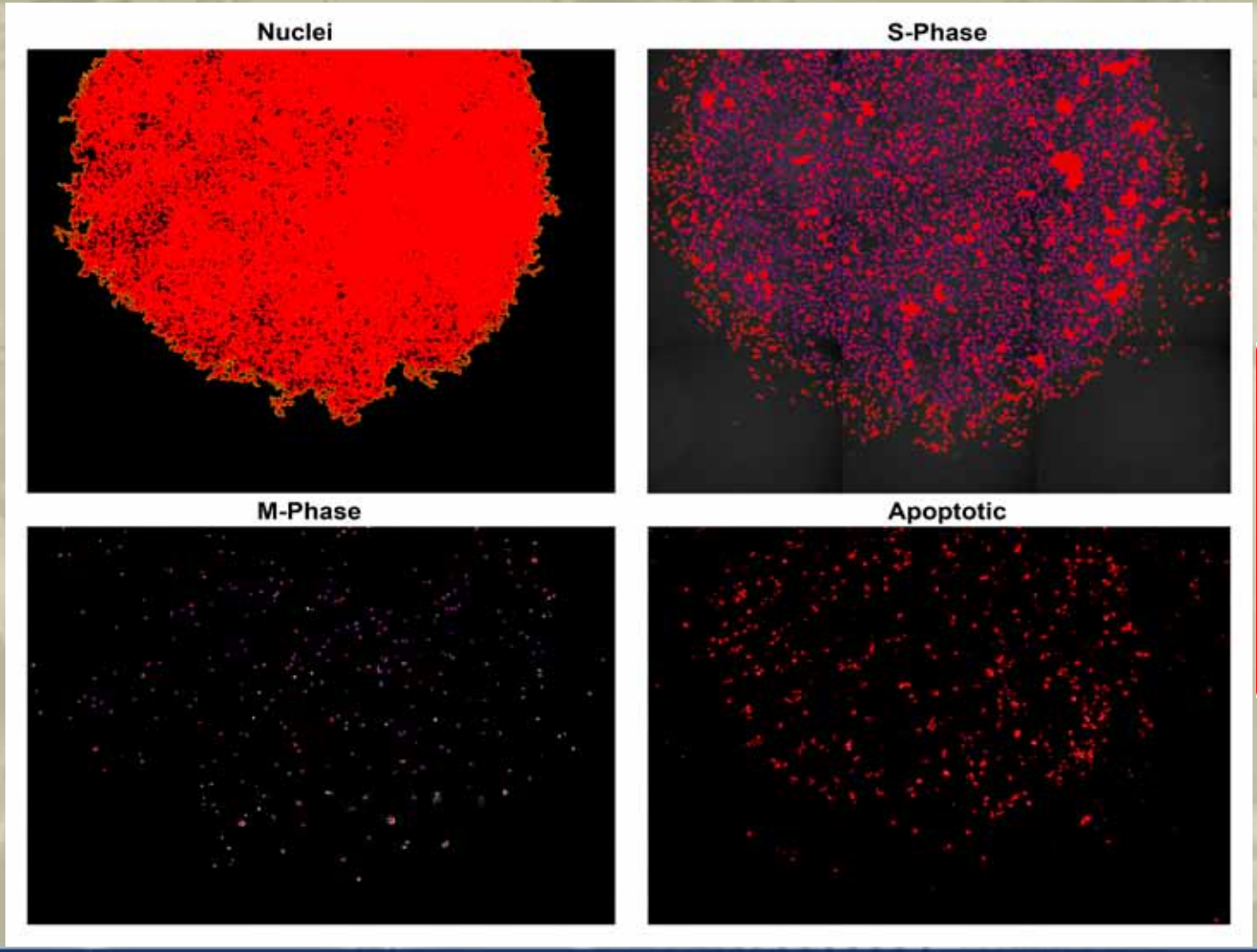


Signal Transduction Marker (NFkB), intensity based measurement of biology

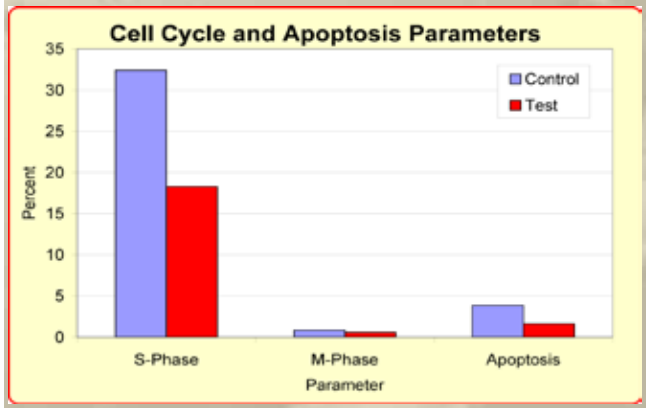


Looks like a biology difference, really a cell cycle difference

BD Pathway: Examples for Applications : Image and Data Analysis of hES Colony



Area based measurements



BD

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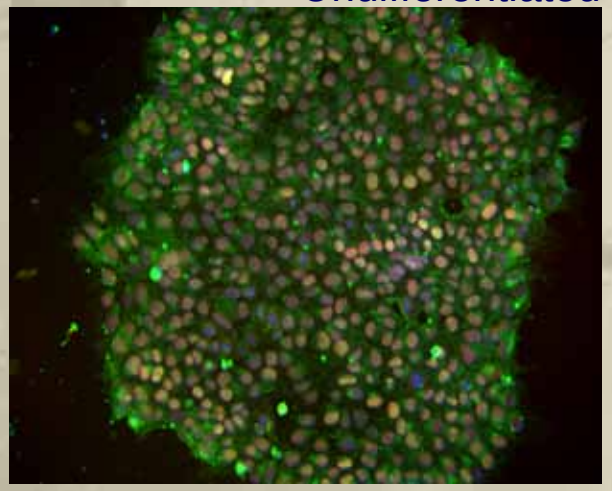
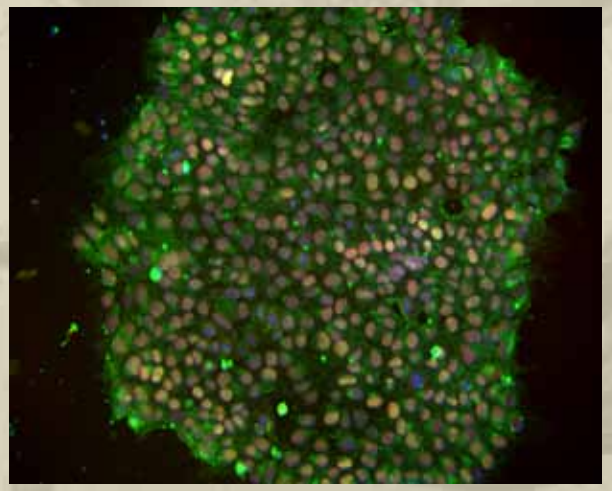
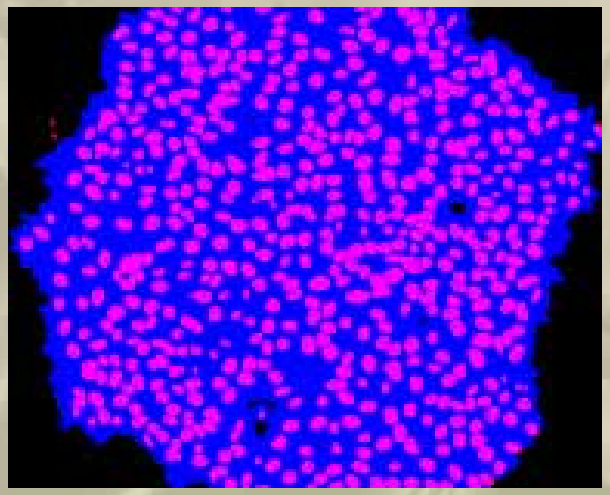
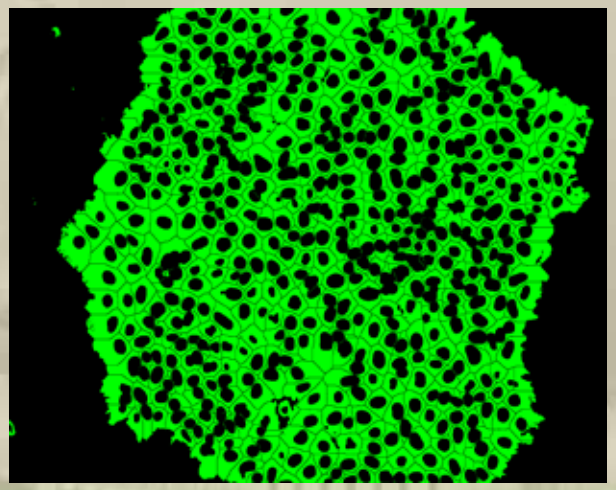
BD Pathway: Examples for Applications : Image and Data Analysis of hES Colony Representative Segmentation Masks

Undifferentiated hES cells

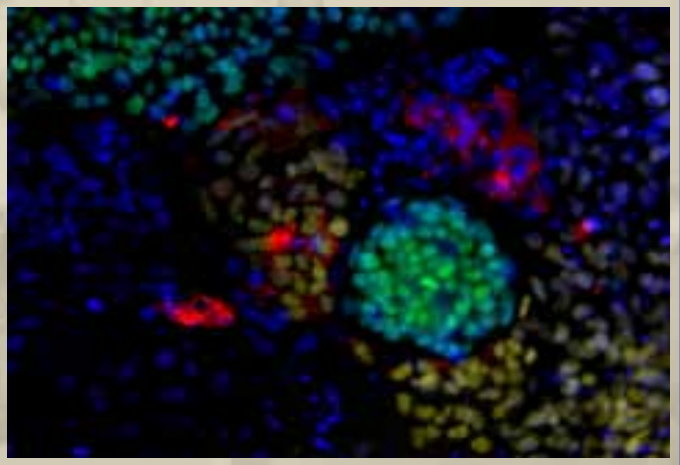


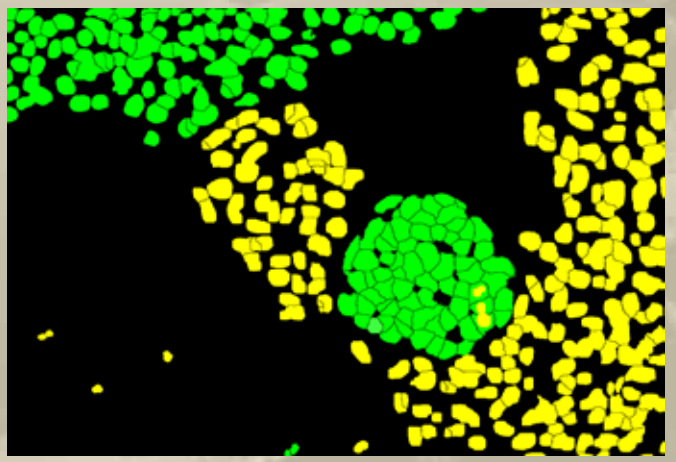






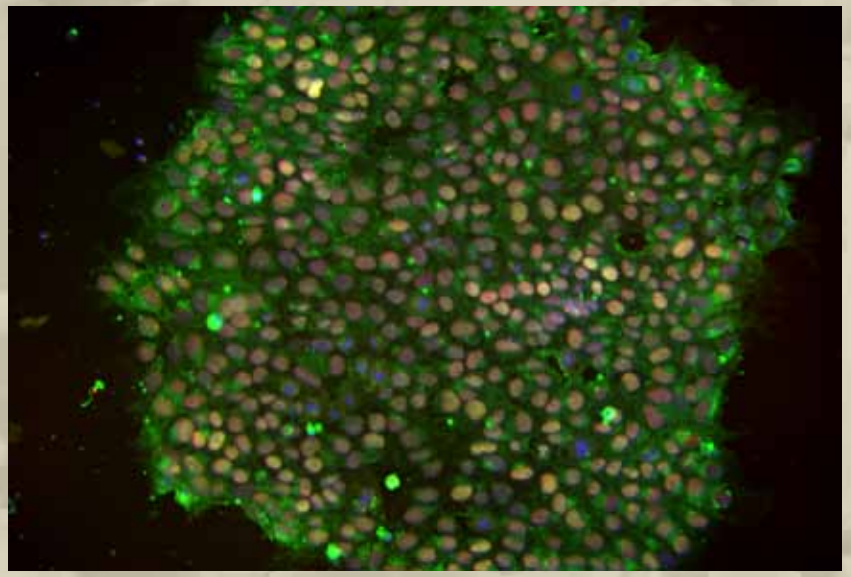
Differentiated hES cells



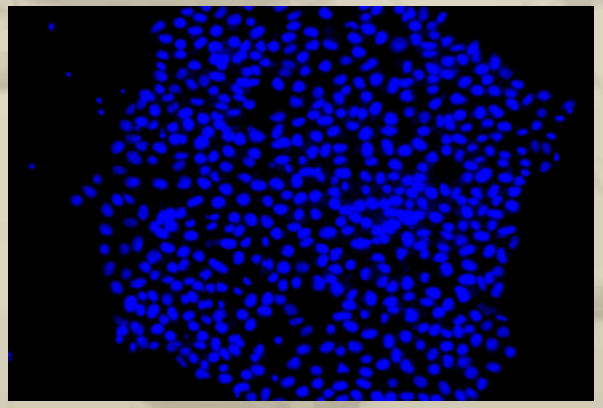


BD Pathway: Examples for Applications : Image and Data Analysis of hES Colony Analysis of Differentiation - Undifferentiated

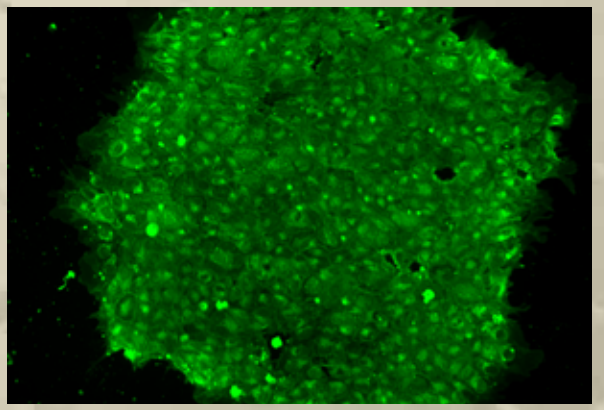
Merged image



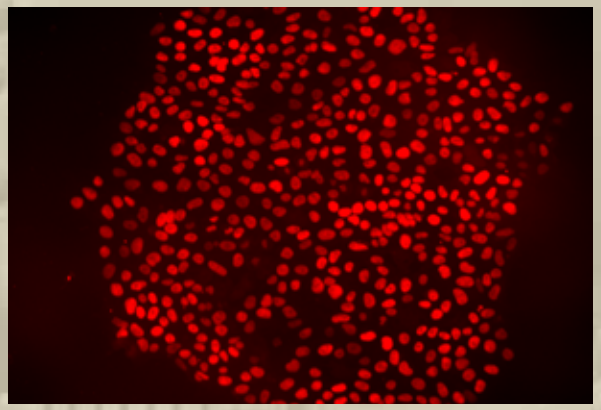
Hoechst



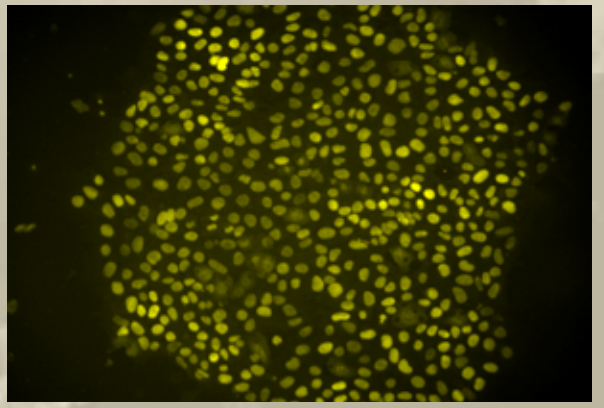
SSEA-4 (- diff)

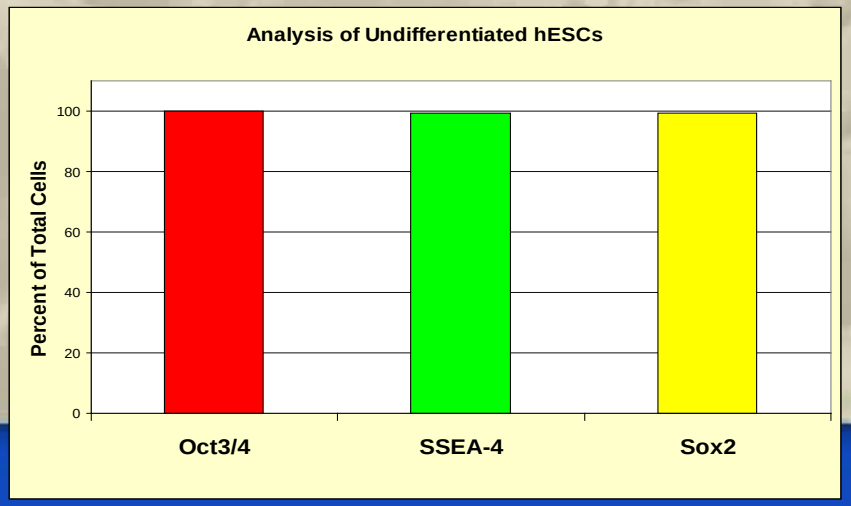


Oct 3/4 (- diff)



Sox2 (- diff)





BD

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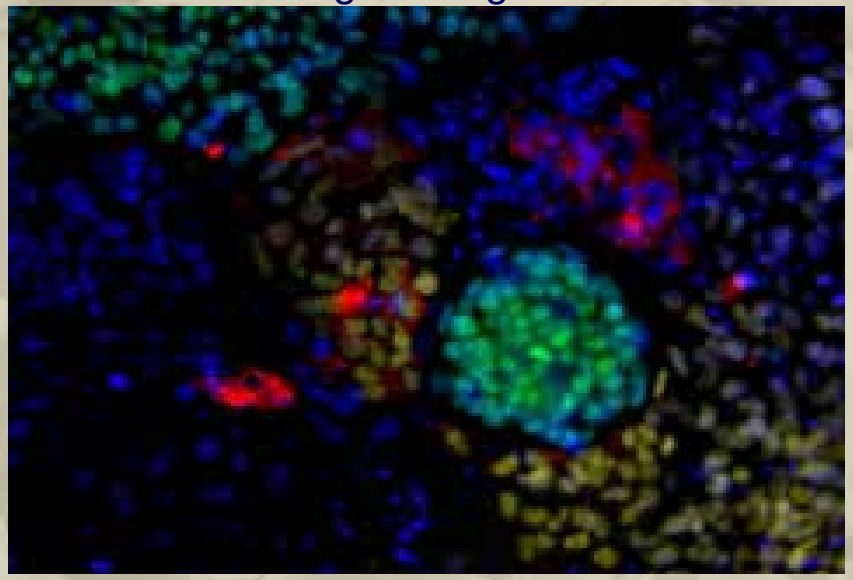
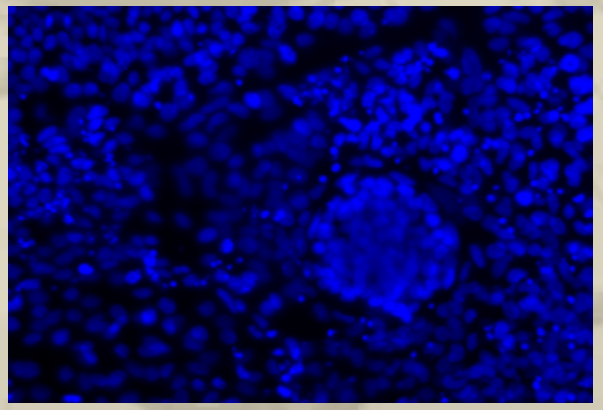
BD Pathway: Examples for Applications : Image and Data Analysis of hES Colony Analysis of Differentiation - Undifferentiated

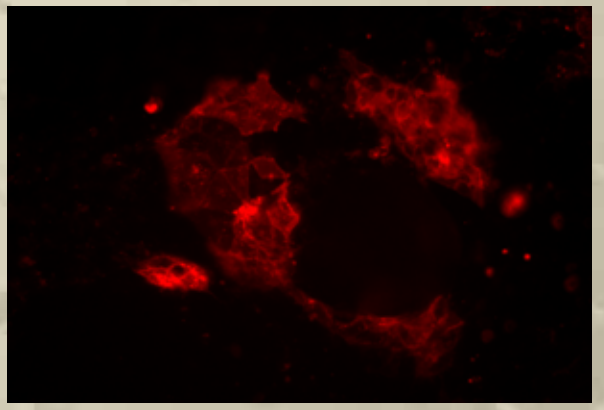
Merged image



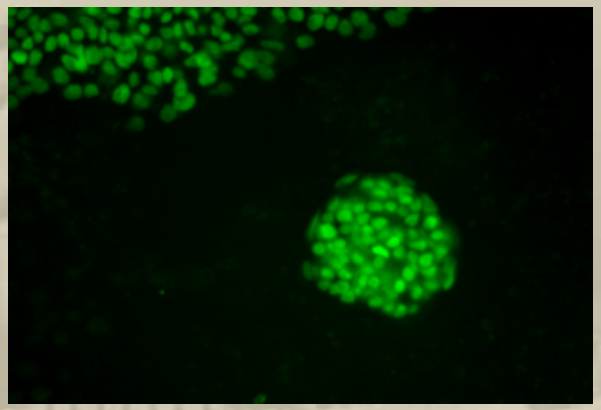
Hoechst



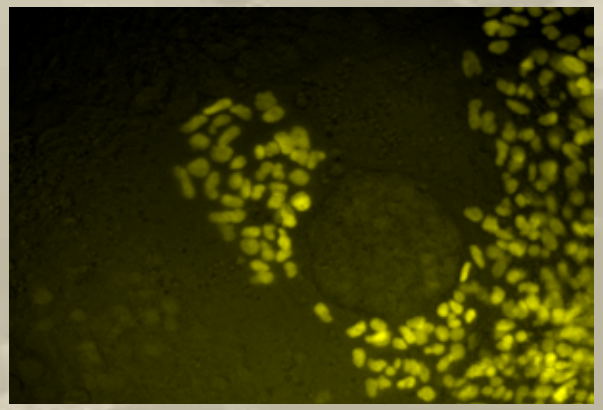
SSEA-1(+ diff)

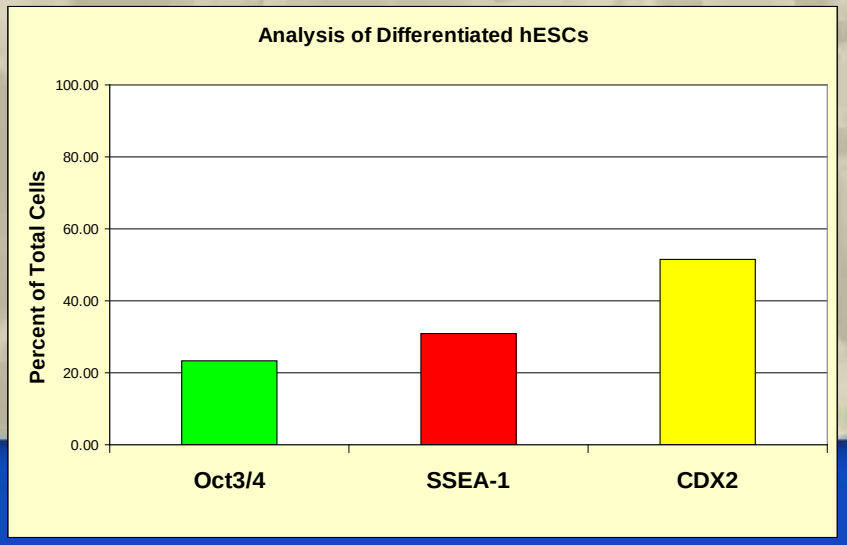


Oct 3/4 (- diff)



CDX2 (+diff)





BD

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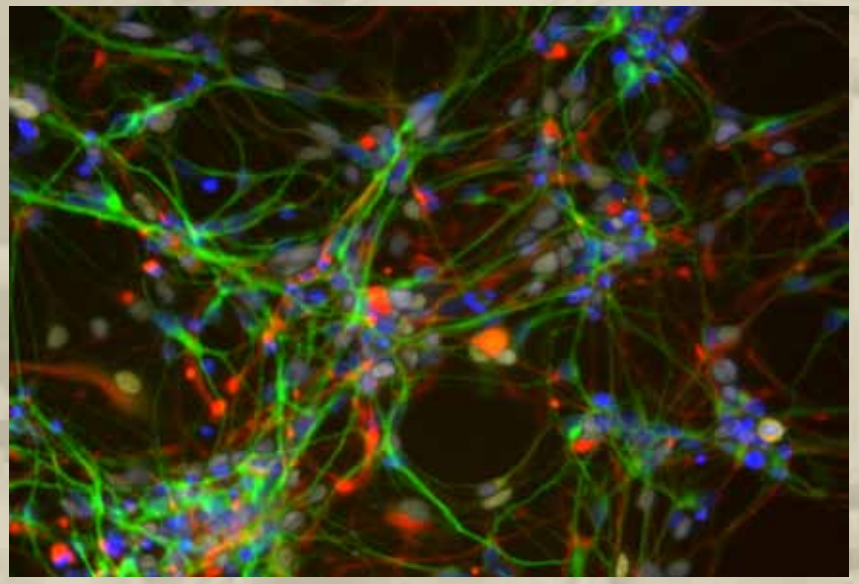
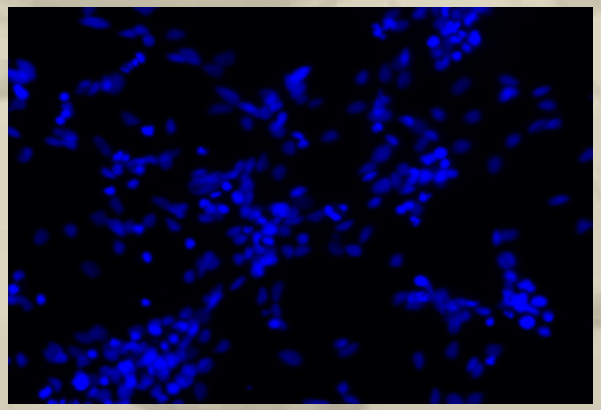
BD Pathway: Examples for Applications : Image and Data Analysis of hES Colony: Pre Sort Images

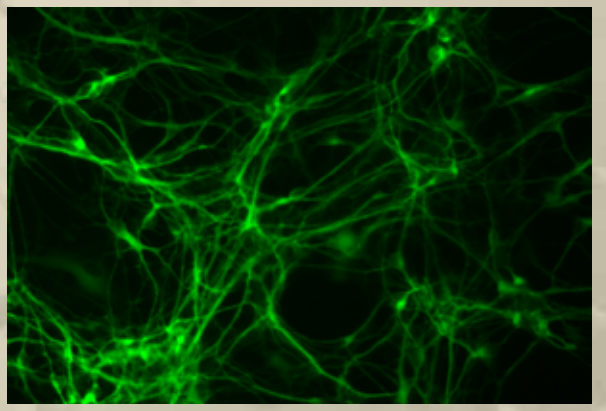
Merged image



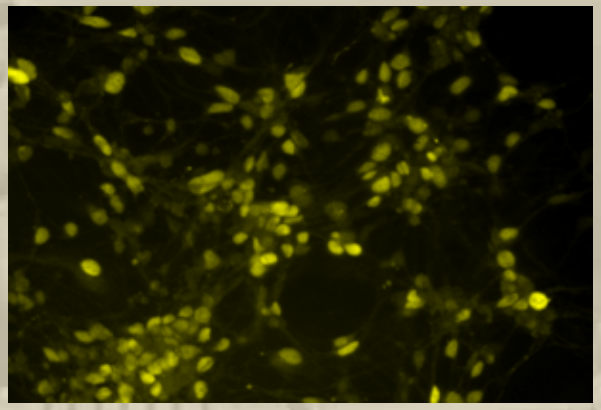
Hoechst



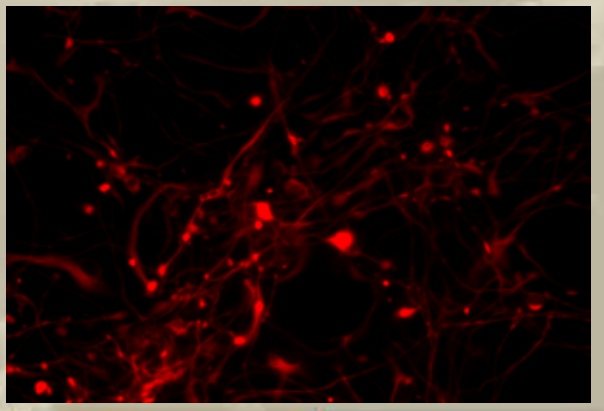
MAP2B (neurons)



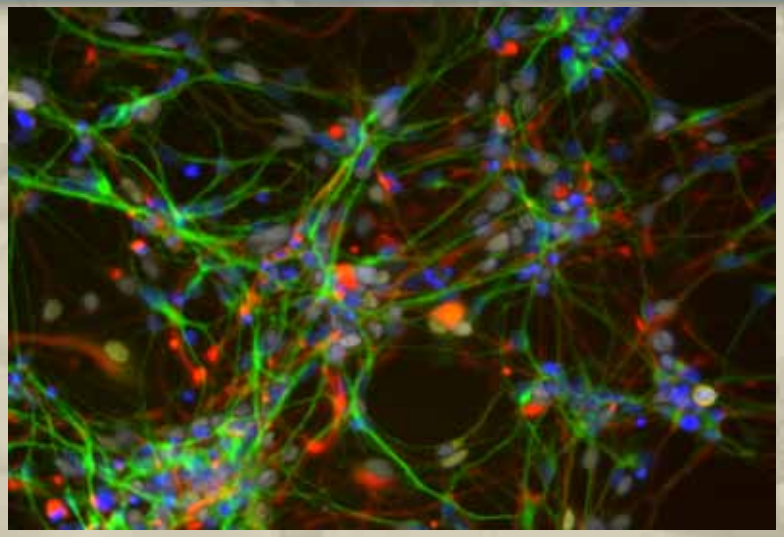
Sox2 (neuronal stem)

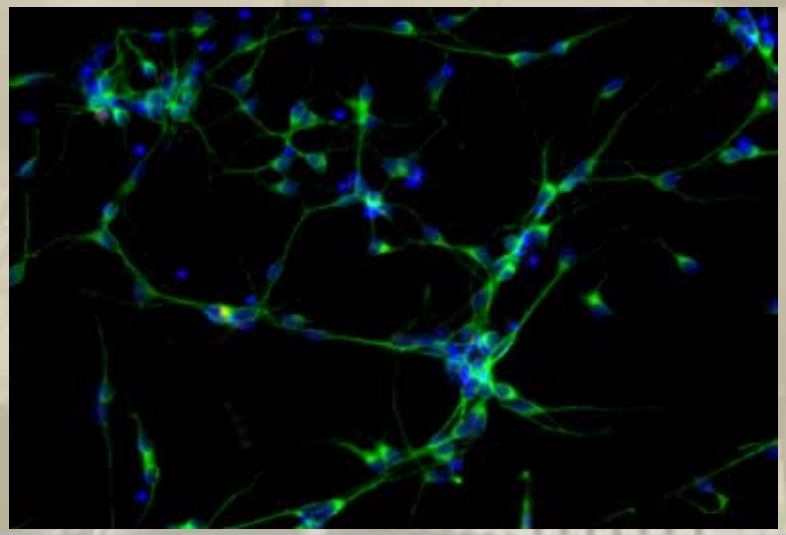


Nestin (neuronal stem)

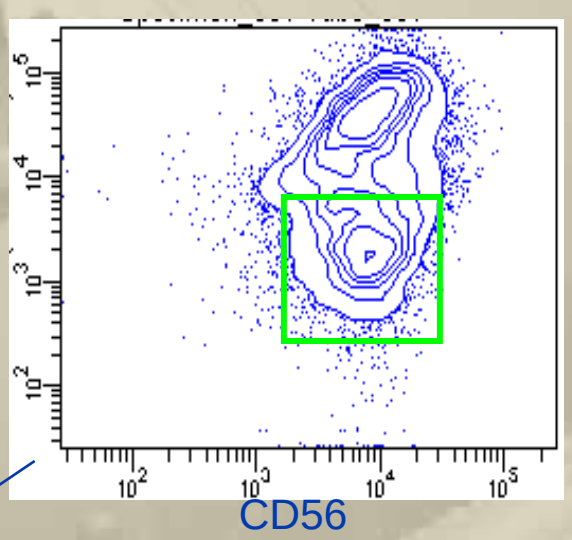


BD Pathway: Examples for Applications : Image and Data Analysis of hES Colony Differentiation of Neuronal Stem Cells





hESCs differentiated into neurons produce a mixed culture. Cells were flow sorted to isolate the $CD56^{+}/CD184^{dim}$ population



Sorted population cultured for additional 10 – 14 days

BD

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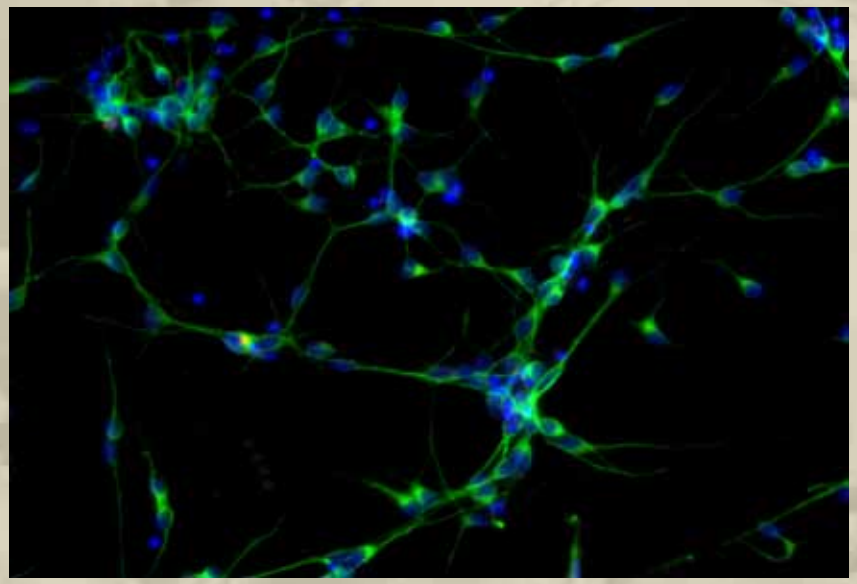
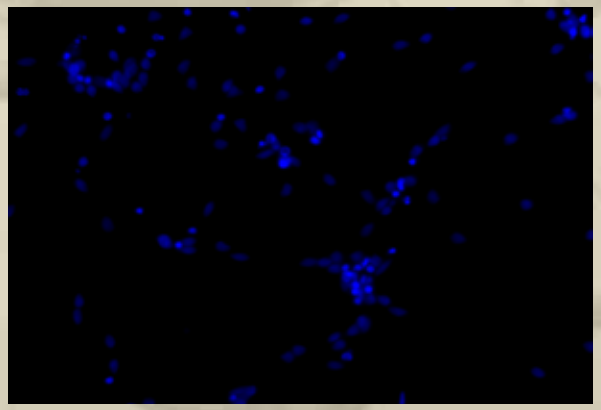
BD Pathway: Examples for Applications : Image and Data Analysis of hES Colony Post Sort Images and Analysis

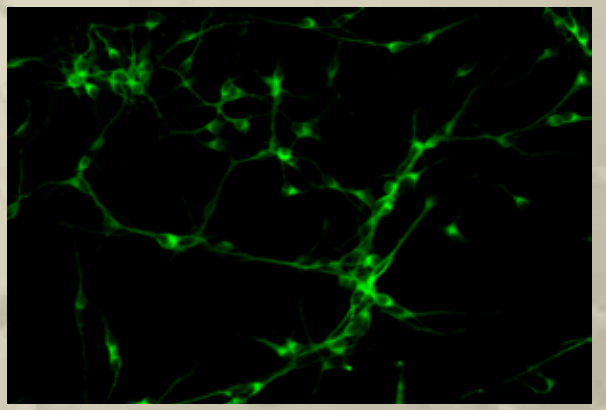
Merged image



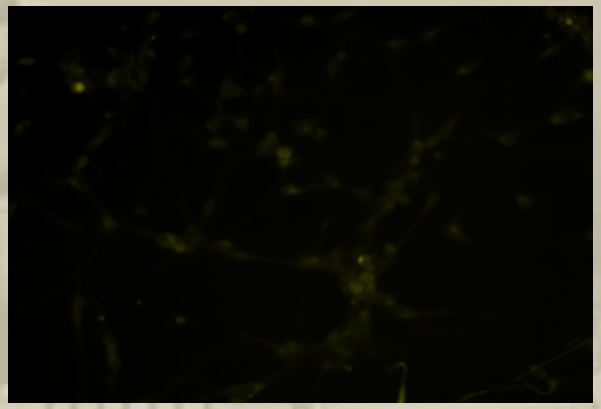
Hoechst



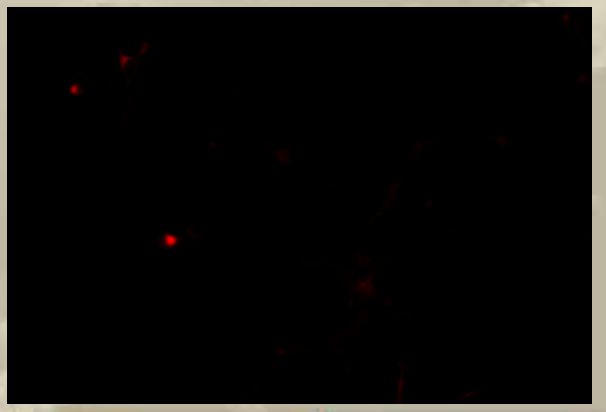
MAP2B (neurons)



Sox2 (neuronal stem)

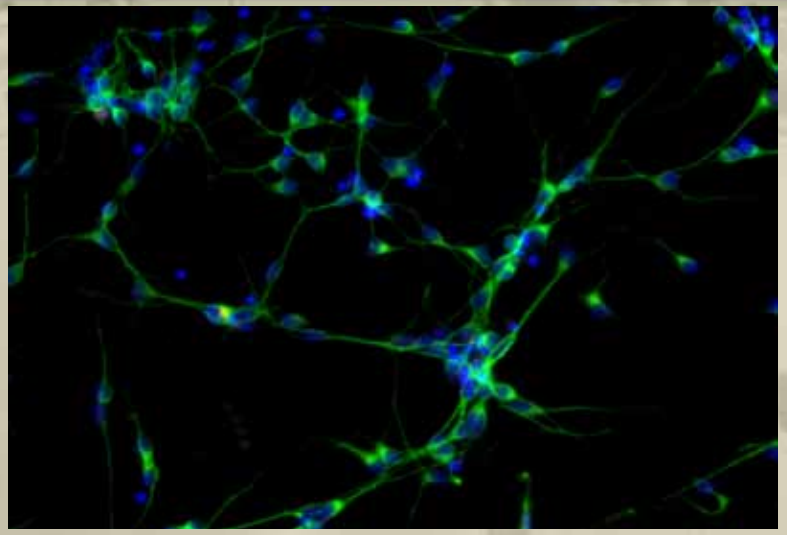


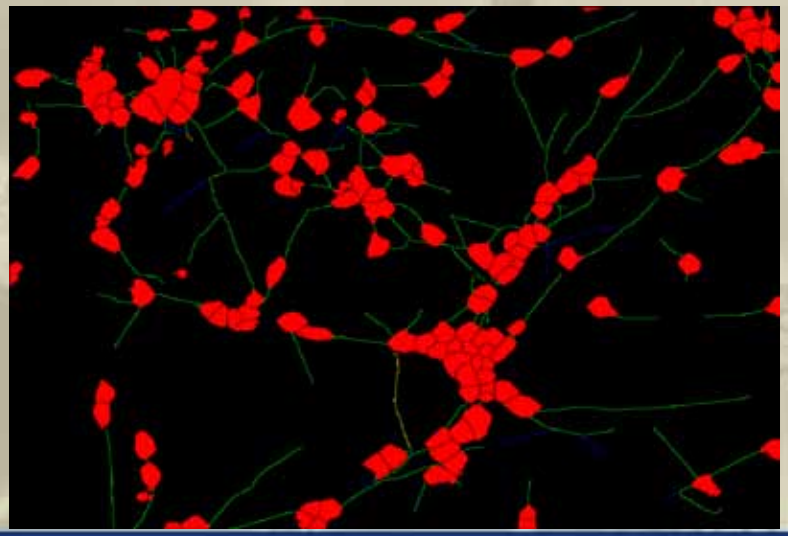
Nestin (neuronal stem)



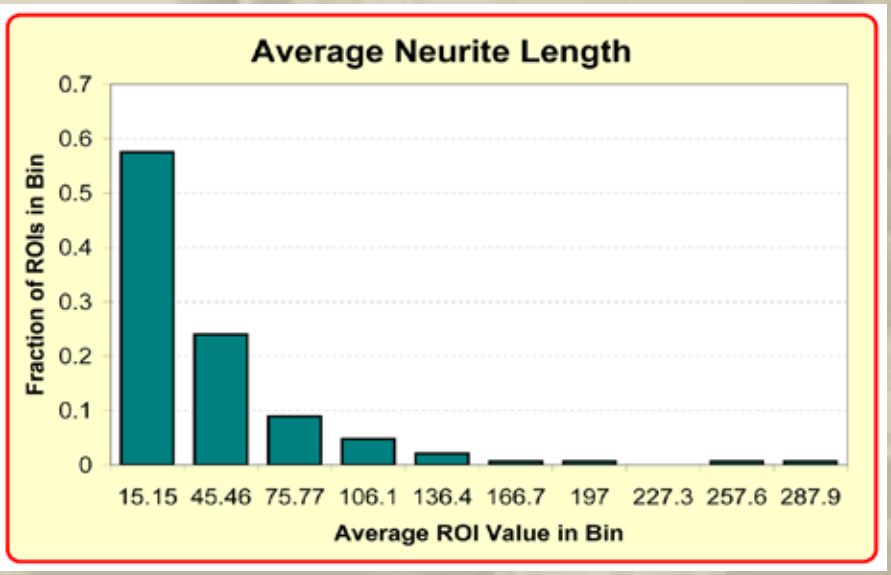
	Sox2	Nestin	Map2B	Hoechst
Post Sort	16	9	123	134
Percent	11.9	6.7	91.8	100

BD Pathway: Examples for Applications Neurite Outgrowth Analysis in hES





Neurites can be segmented and measured using Neurite Outgrowth software



BD

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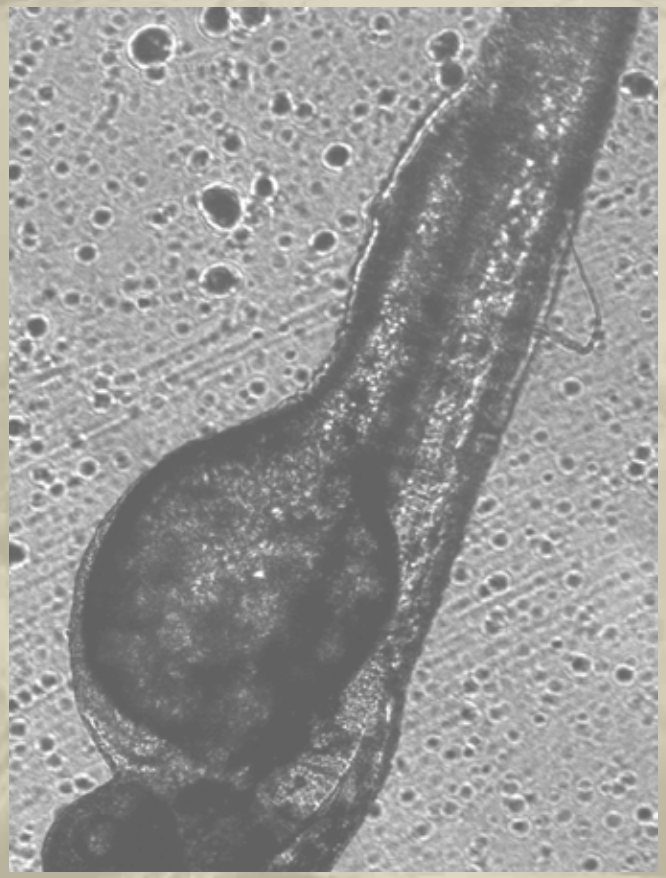
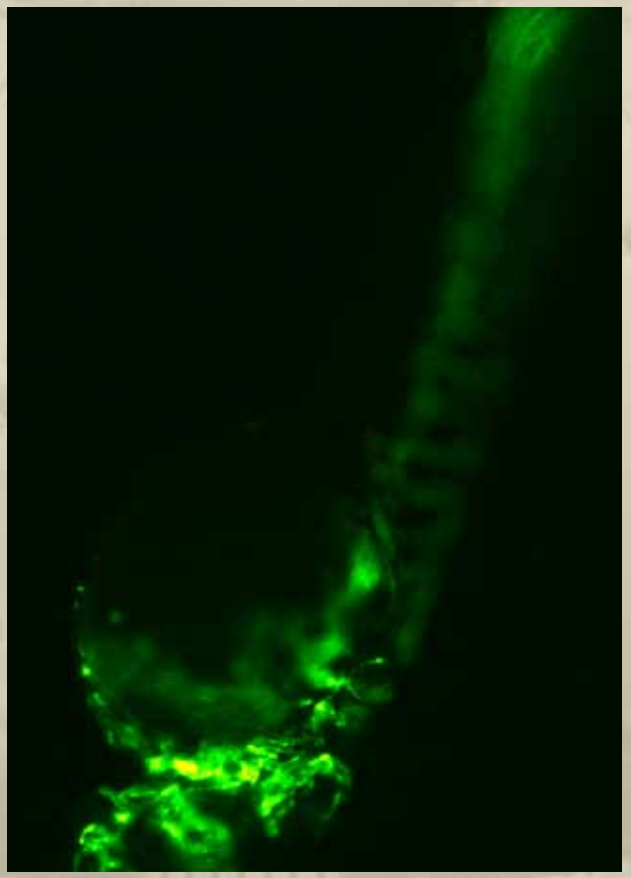
BD Pathway 855: Examples for Applications
Angiogenesis in whole organisms
Confocal Imaging of Zebrafish expressing GFP

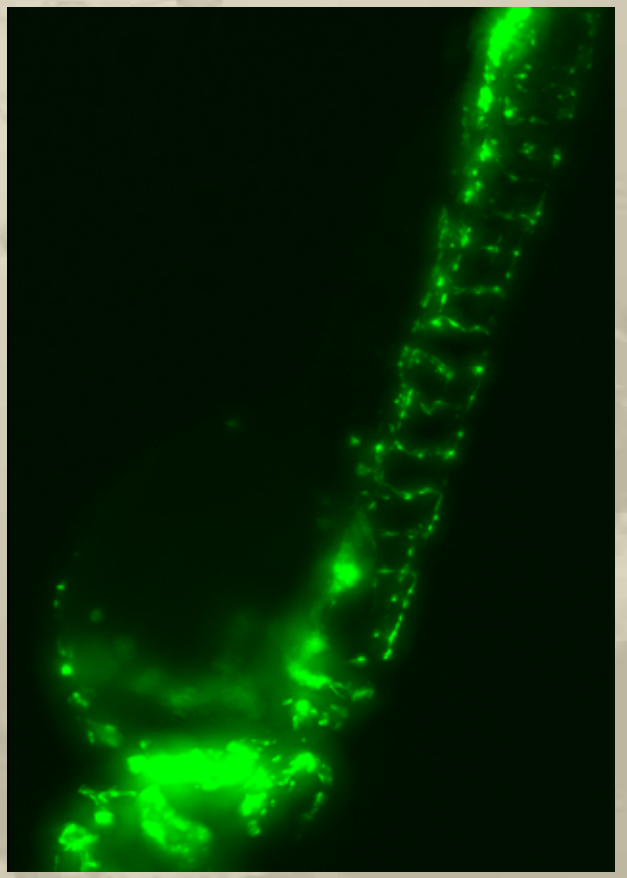
Angiogenesis-GFP Control



Time-lapse, Transmitted



Confocal Z-sections



3D Reconstruction

Imaged on Pathway HT: GFP Confocal, 384-well plate

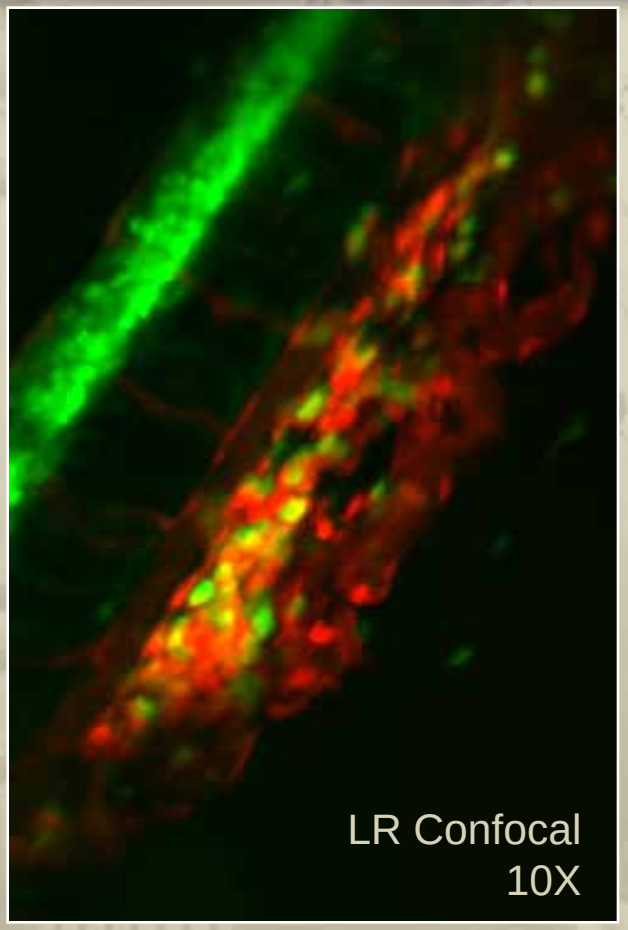
BD Pathway: Examples for Applications
Whole organisms: Zebra Fish

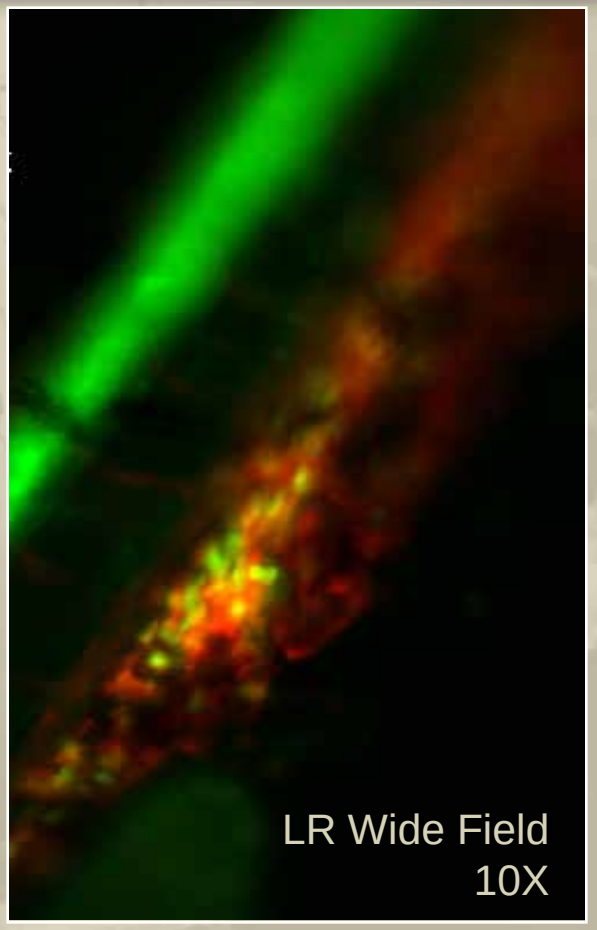
Stunned Fish Placed on Slides

10X Objective
Wide Field Imaging
3X3 montage

10X Confocal
3X3 montage
1.5um Stacks
11 Sections

Cells should always be positive for DsRed
And only sometimes positive for GFP
Those cells that are positive for both will be
Blood Stem Cells (Yellow)



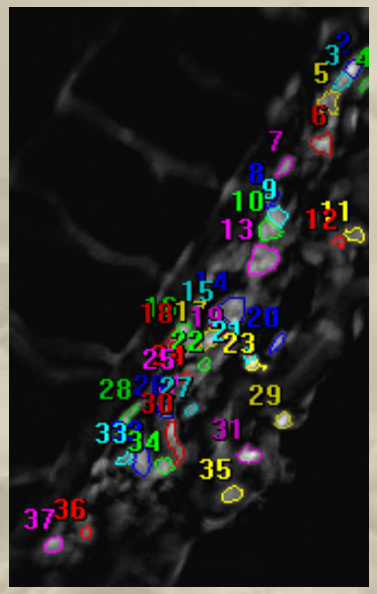


GFP: 100ms
DsRed: 97ms

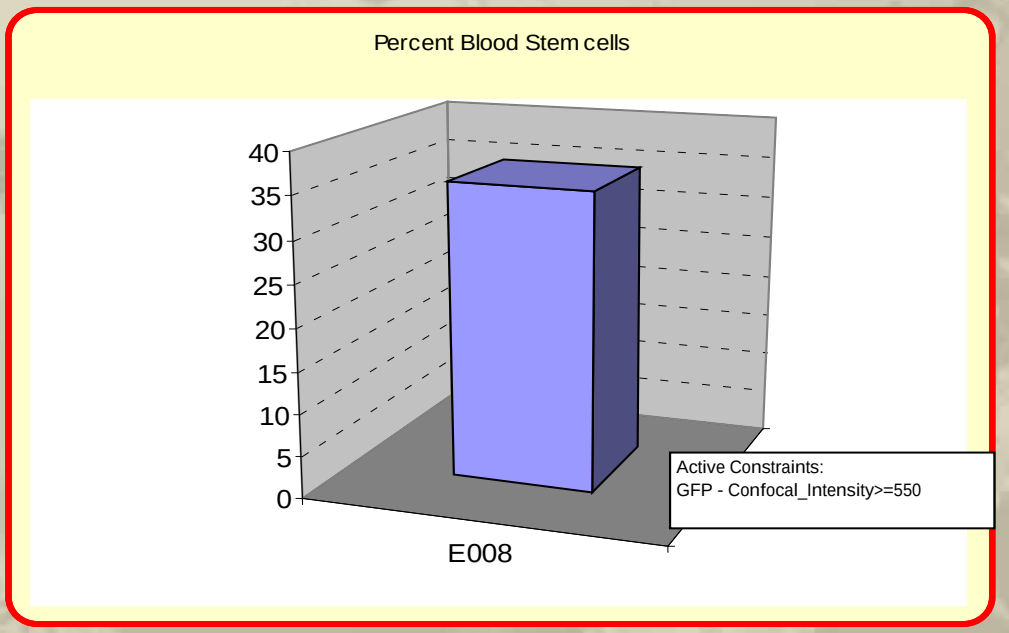
GFP Confocal: 735ms; Gain of 10
DsRed Confocal: 1.7 sec

BD Pathway: Examples for Applications
Whole organisms: Zebra Fish
Collapsed Stack Confocal Image Analysis

Rolling Ball Background Subtraction on DsRed Image
Polygon Segmentation using watershed to split cells
Measure DsRed and GFP Intensity with Nuclear Mask



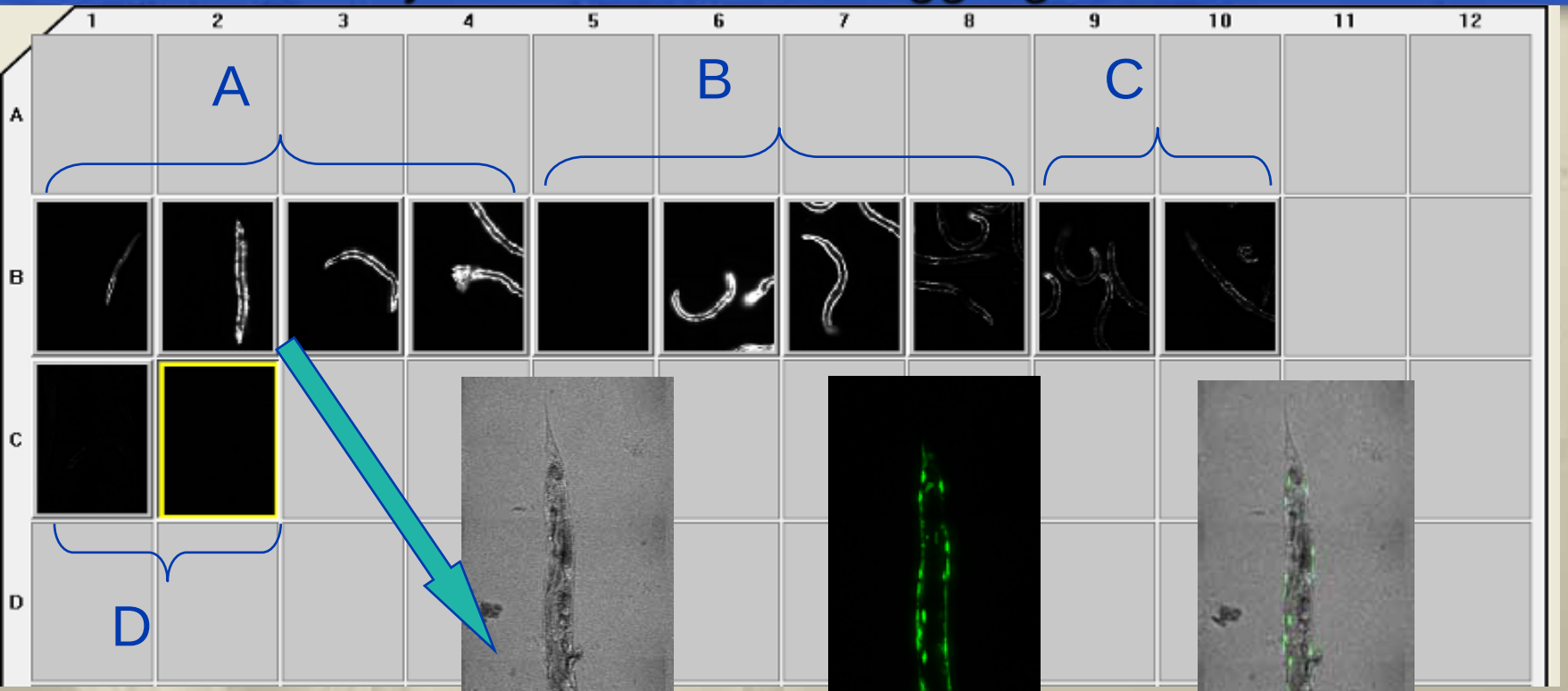
LR Confocal



Data Constrained: GFP>550

Comments: Images and analysis would be improved with 20X confocal

BD Pathway: Examples for Applications
Whole organisms: C. elegans
Analysis of fluorescent aggregates in 96 Well Plate



Long Exposure,
200-3000 dynamic range

BD

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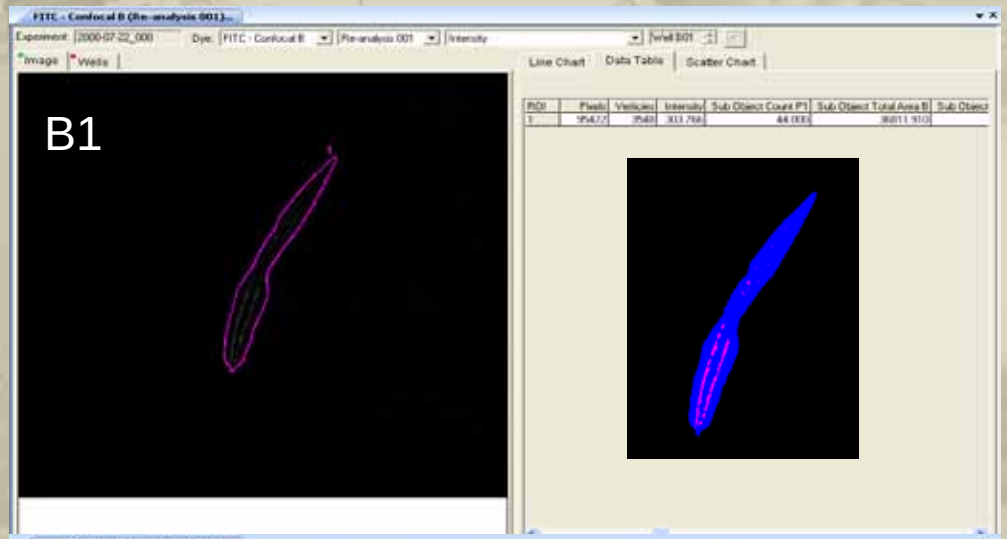
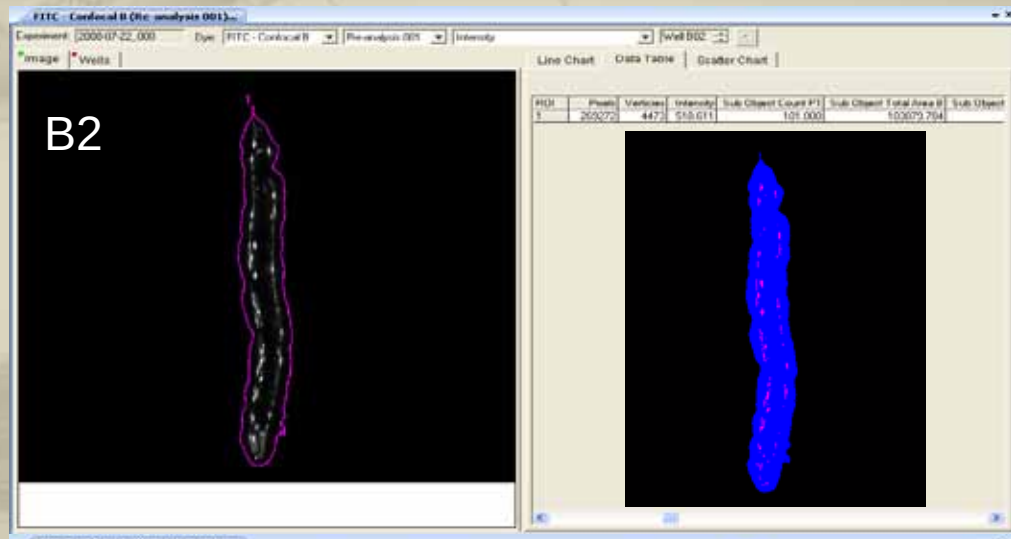
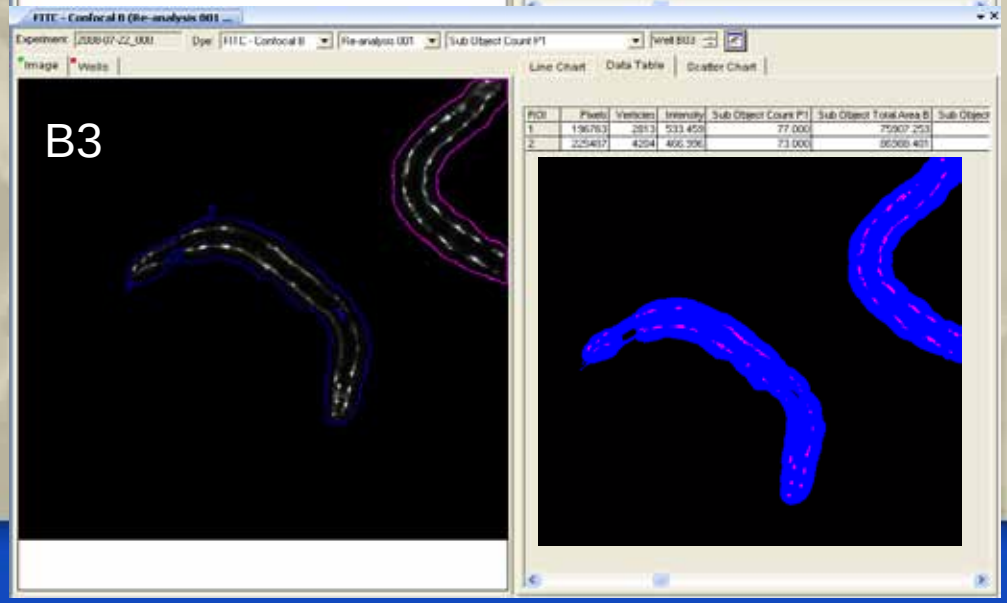
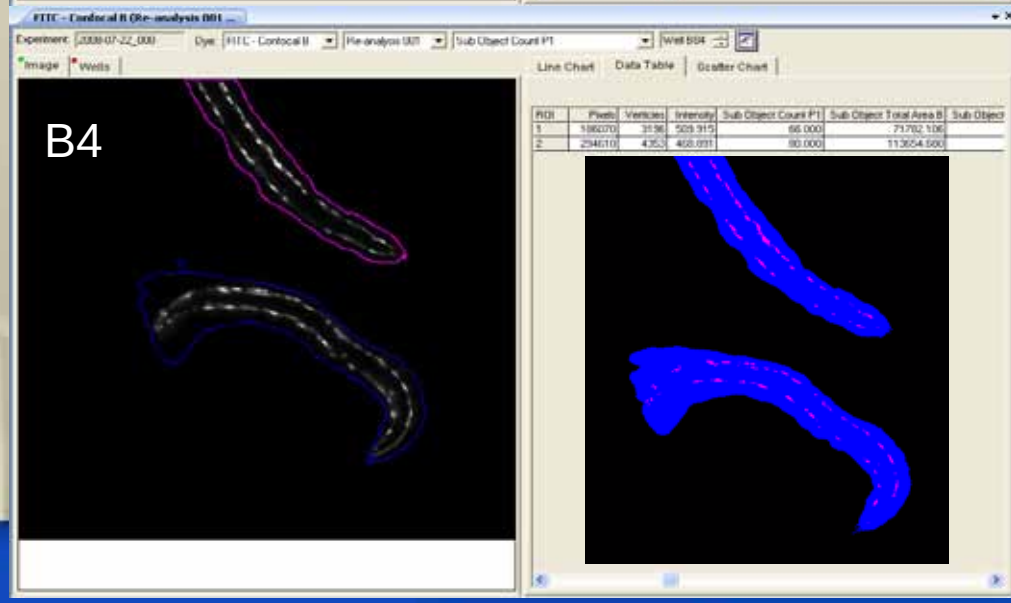
BD Pathway: Examples for Applications
Whole organisms: C. elegans
Analysis of fluorescent aggregates



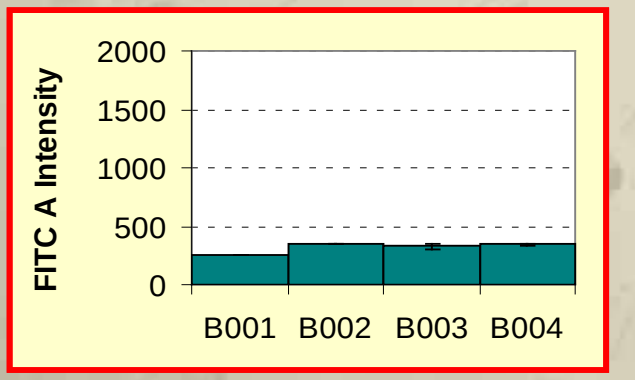




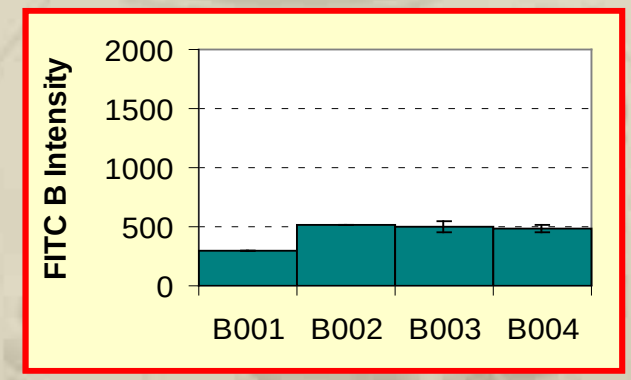


BD Pathway: Examples for Applications
Whole organisms: C. elegans
Analysis of fluorescent aggregates

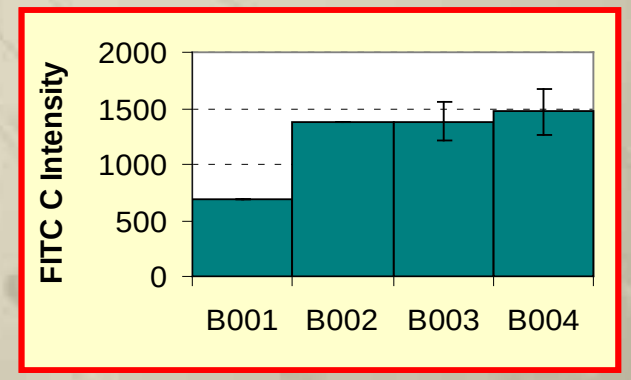
10 ms



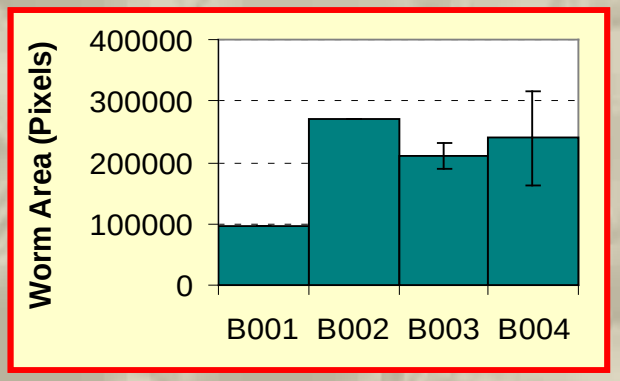
25 ms



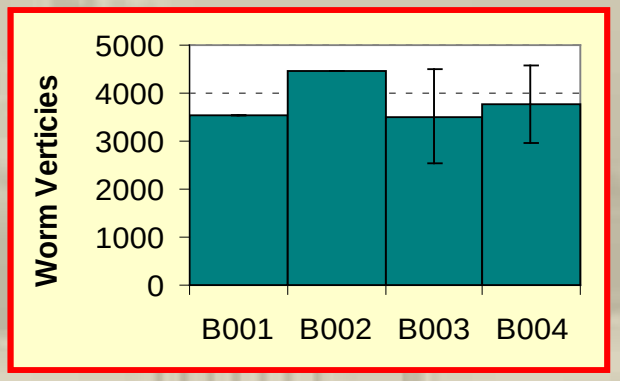
100 ms

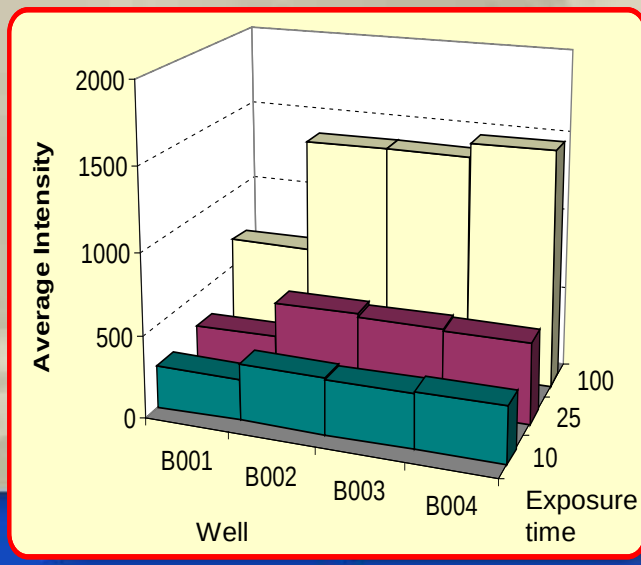


Area



Vertices

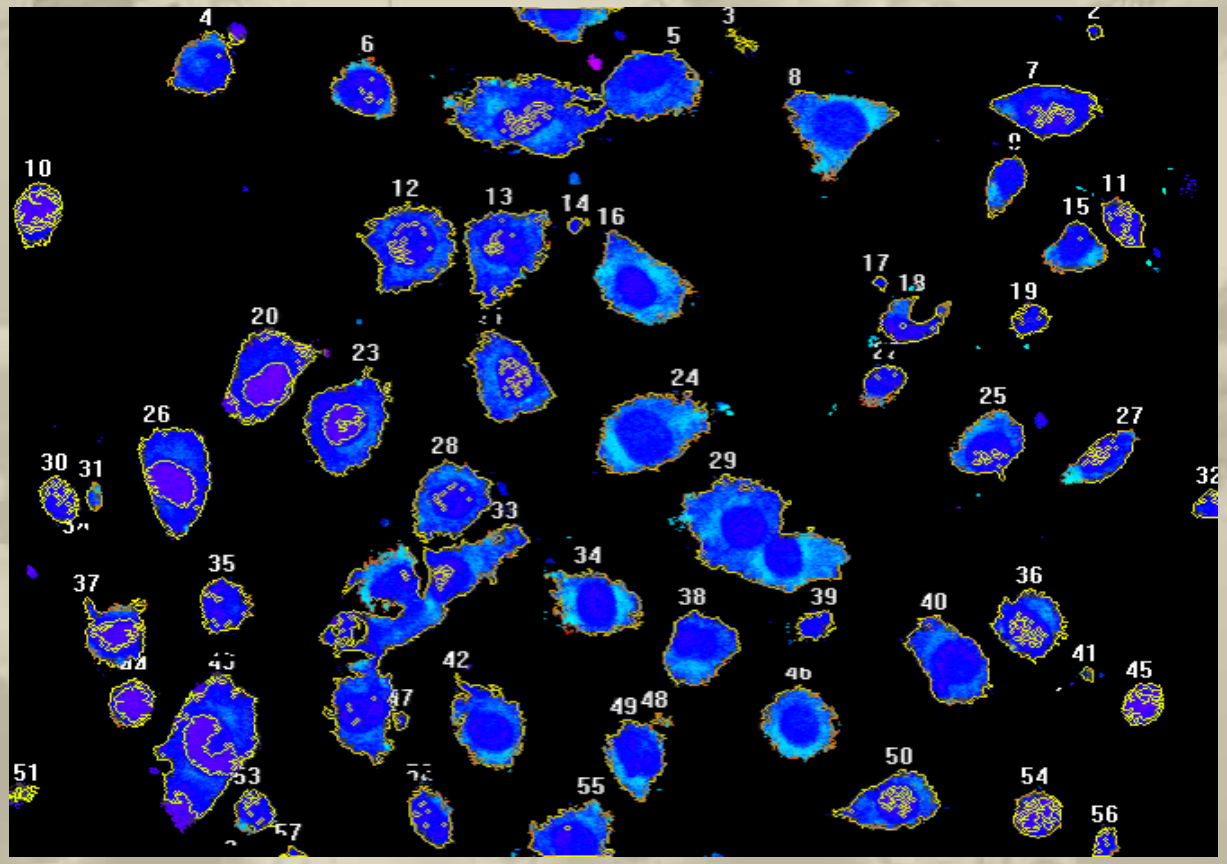




BD Pathway 855:
Examples for Applications

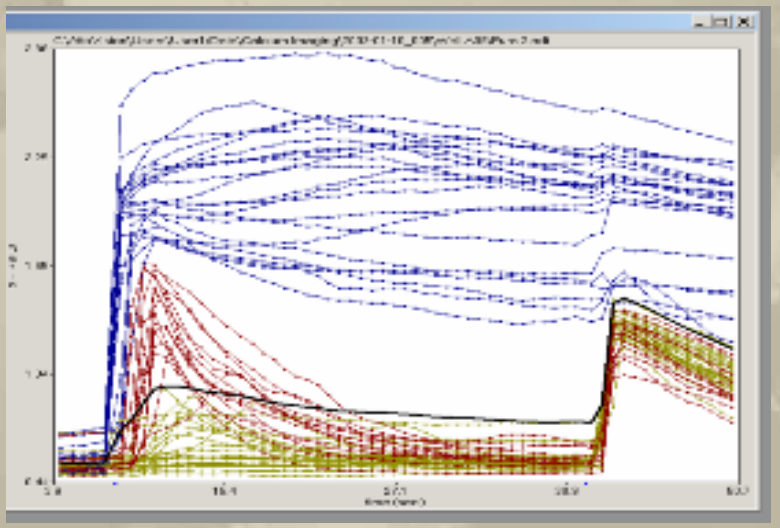
3. 2. Kinetic Assays

BD Pathway 855: Examples for Applications
Whole organisms: Fura-2 ratiometric calcium imaging



Live Cell
Kinetic Calcium Assay

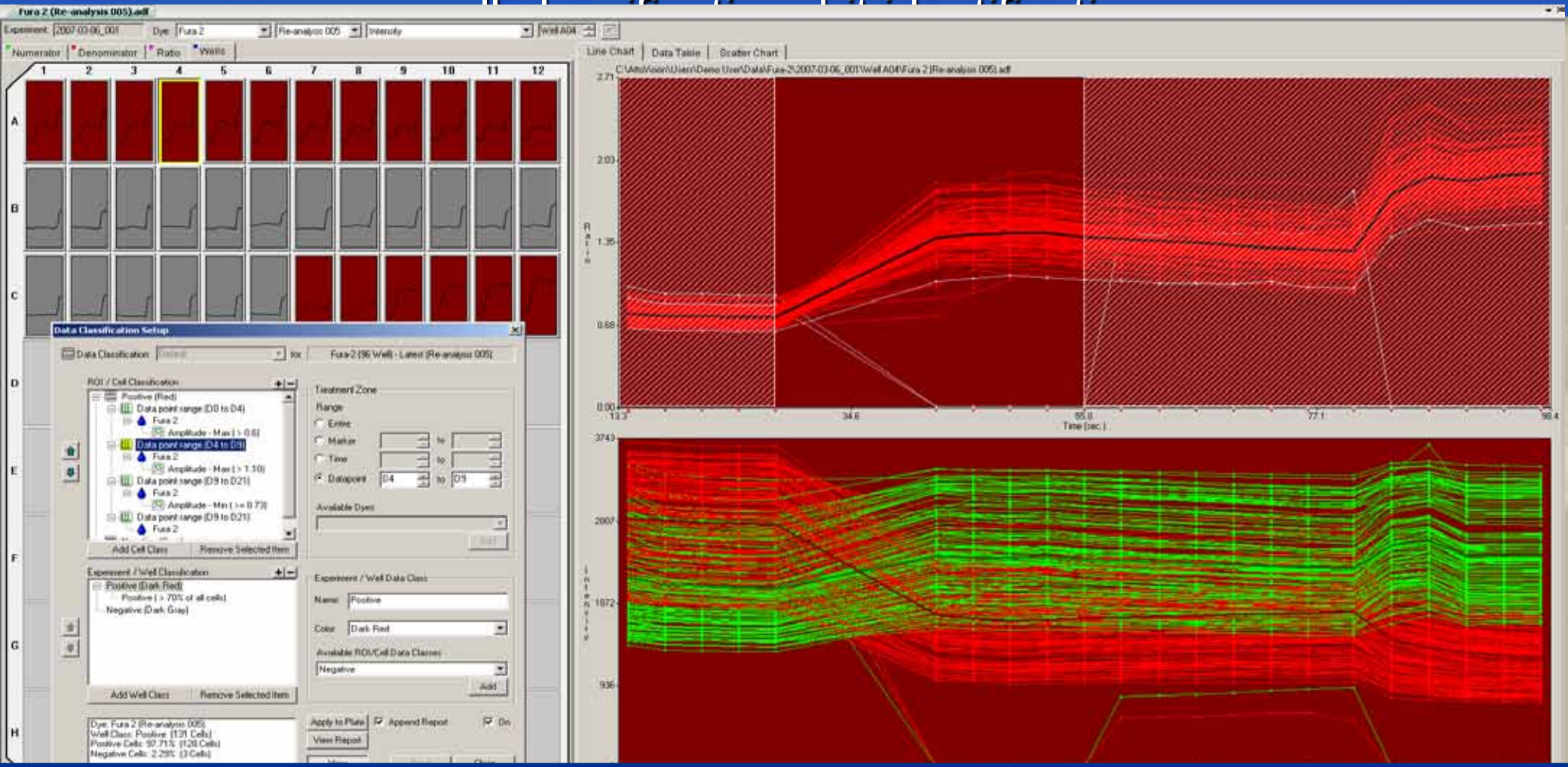
20x objective



Ca^{2+} response in stably transfected (VR1 receptor) NIH 3T3 cells after stimulation with an agonist

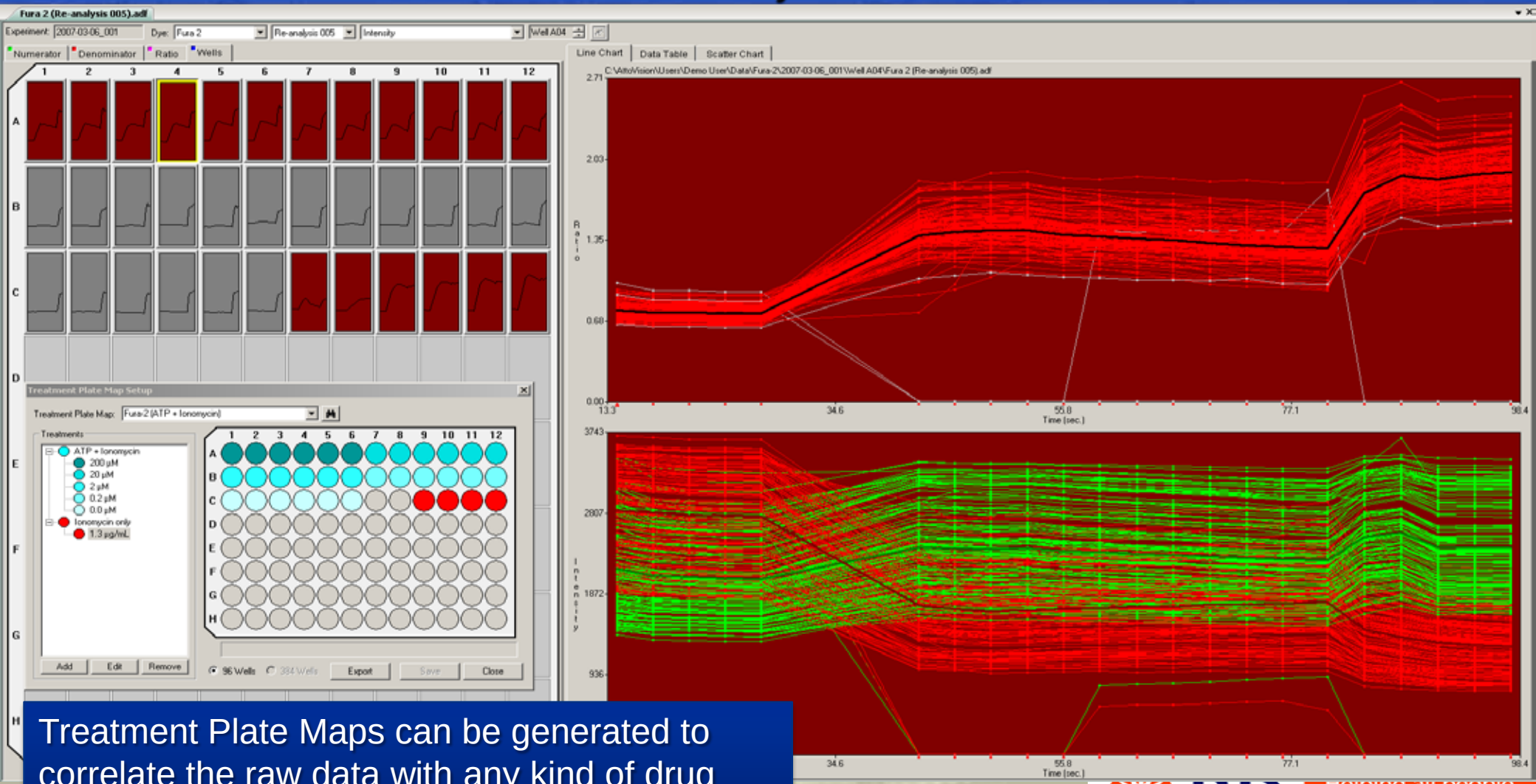
Cells line:Dr. Mike Iadarola, NIDCR, NIH, Bethesda, MD

BD Pathway 855: Examples for Applications
BD Attovision: Data Classification of kinetic calcium assay (Fura-2)



The Data Classification tool allows split up the experiment into treatment zones and classify cells and wells e.g. into positive and negative events according to six criteria per parameter and channel, such as the “Amplitude Maximum, Minimum or Average”, the “Area Under the Curve” or the “Rate of Rise” and “Rate of Fall” (here parameter “Intensity” was chosen for the Fura-2 channel). This classification may be applied to the whole plate and a red thumbnails indicate those wells contains cells that meet the criteria. Multiple classes and corresponding colours can be set up.

BD Pathway 855: Examples for Applications
Combination of well classification with dose-response analysis



Treatment Plate Maps can be generated to correlate the raw data with any kind of drug application

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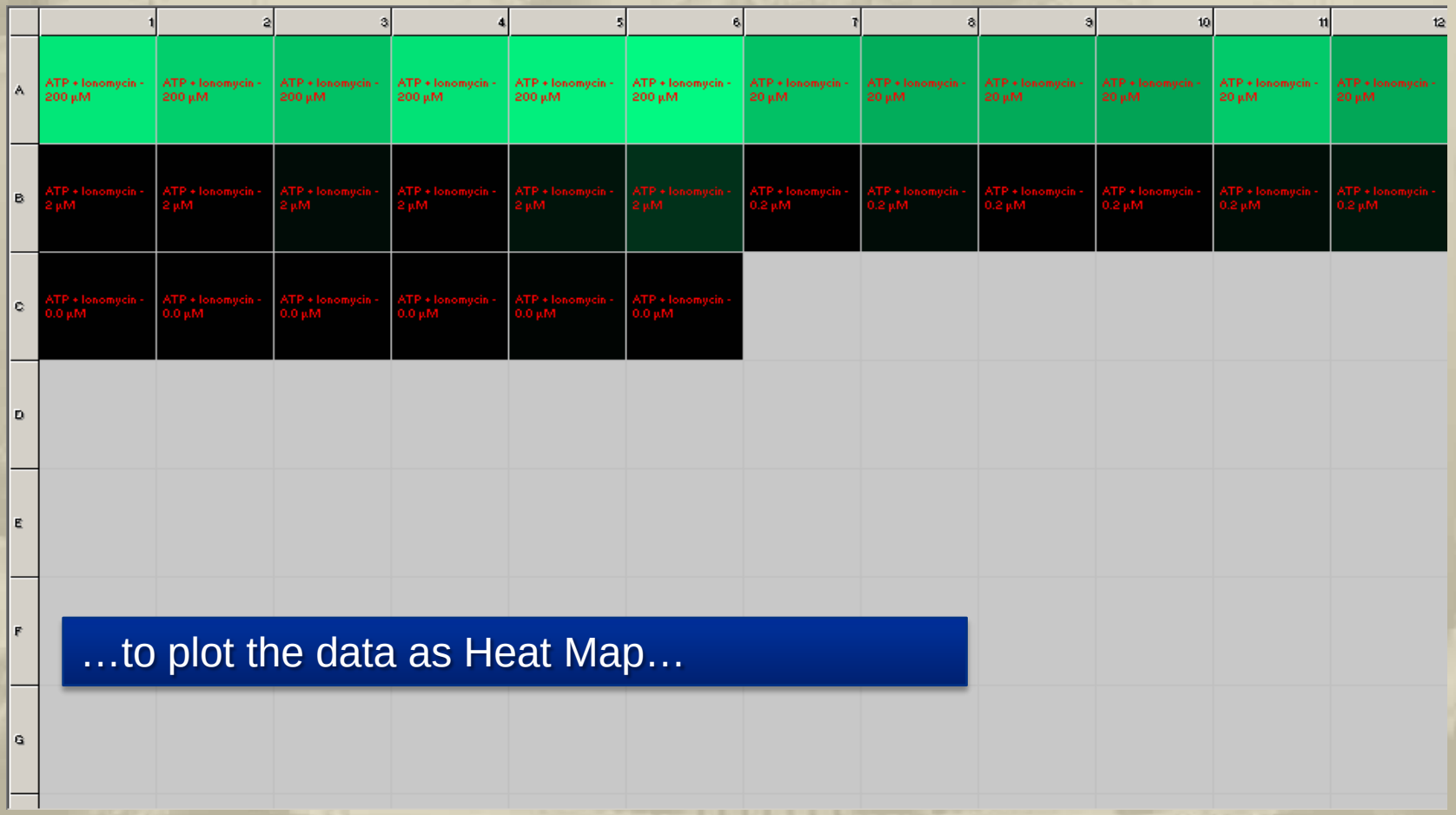
BD Pathway 855: Examples for Applications
Analysis of calcium responses:
Image Data Explorer for data analysis - Heat Maps



…to plot the data as Heat Map…

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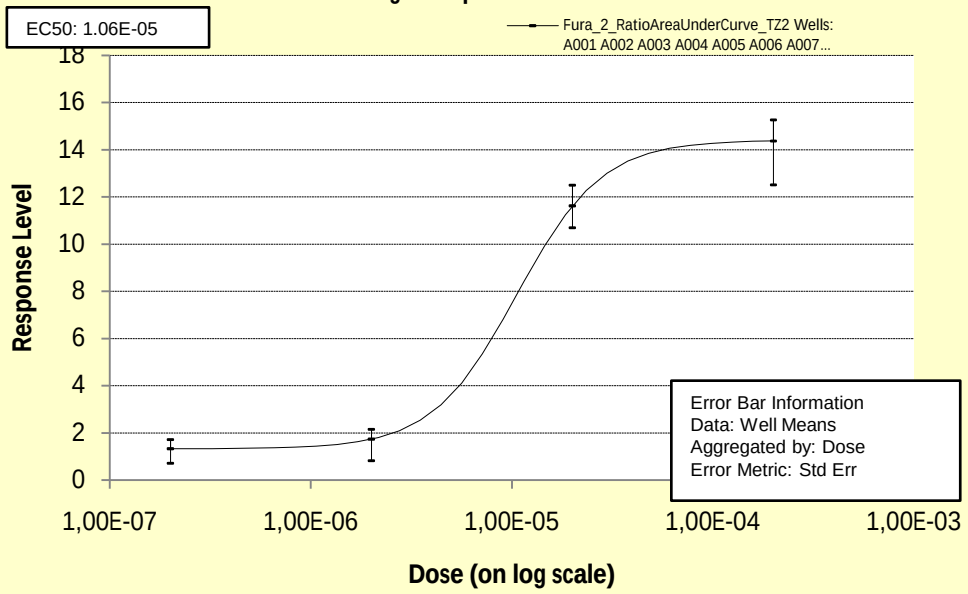
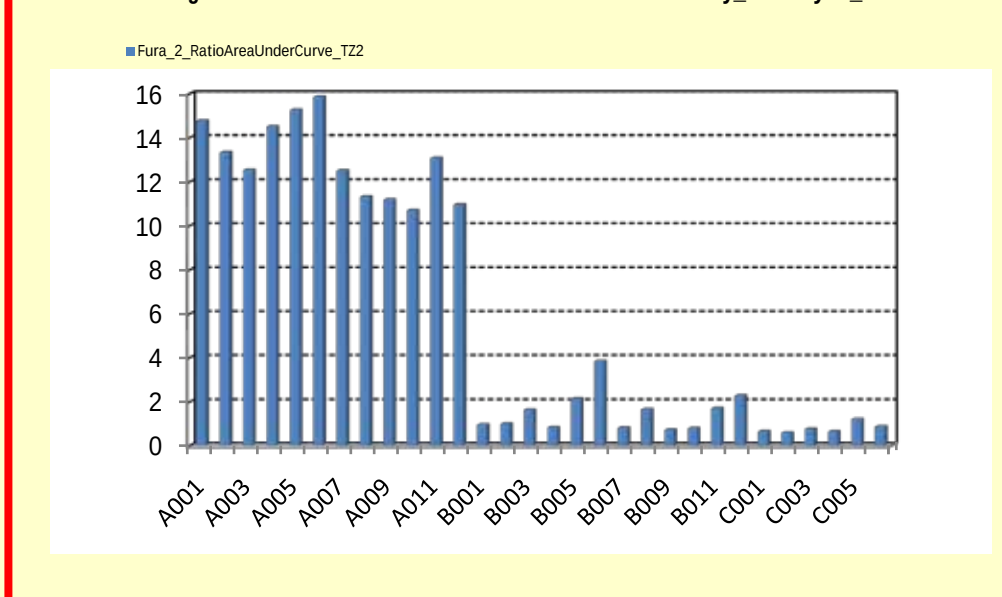
BD Pathway 855: Examples for Applications
Analysis of calcium responses: BD Image Data Explorer for data analysis: graphing the data





…Dose Response Curves or Bar Chart as well as many other graph formats.

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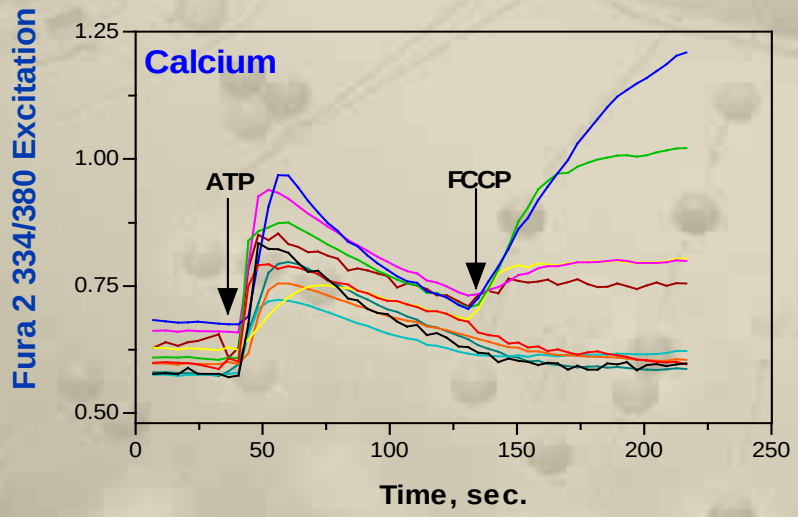
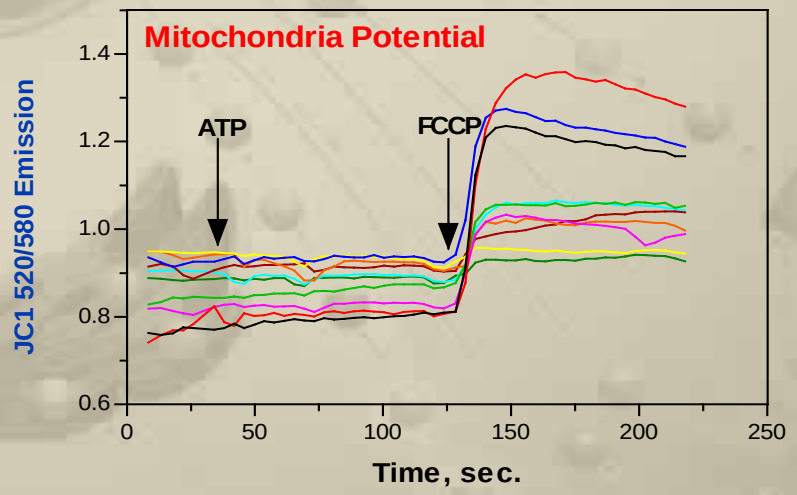
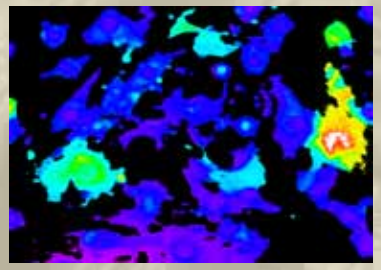
BD Pathway 855: Examples for Applications
Kinetic Multiplexing: Simultaneous Dual Ratiometric Measurement of Calcium and Mitochondria Membrane potential



NIH 3T3 Cells Expressing the Ligand-gated VR1 Ion Channel

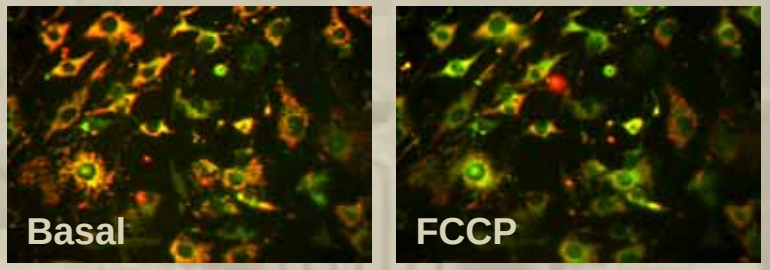


Fura - 2



334/380nm ratio

JC - 1



520nm and 580nm emission images merged

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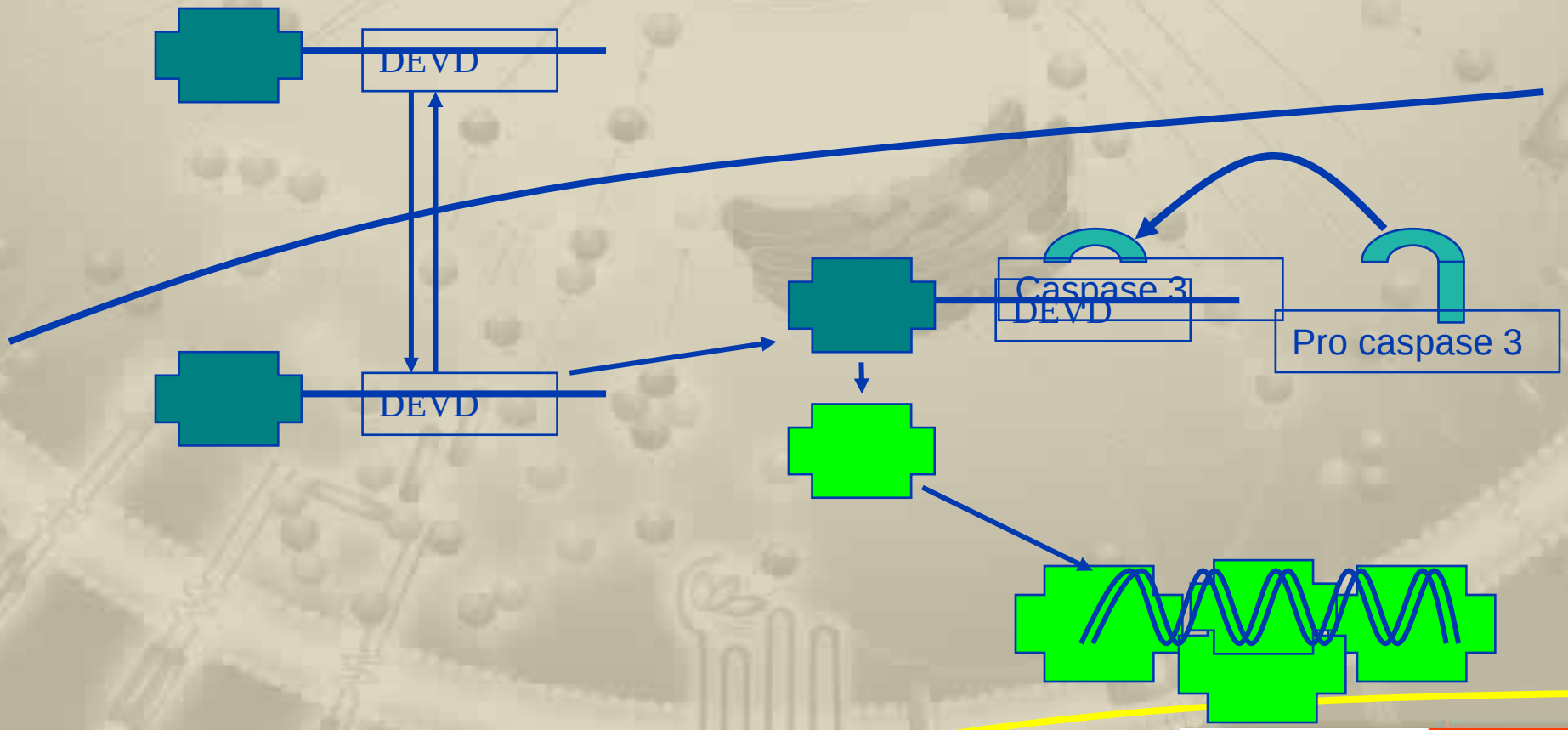
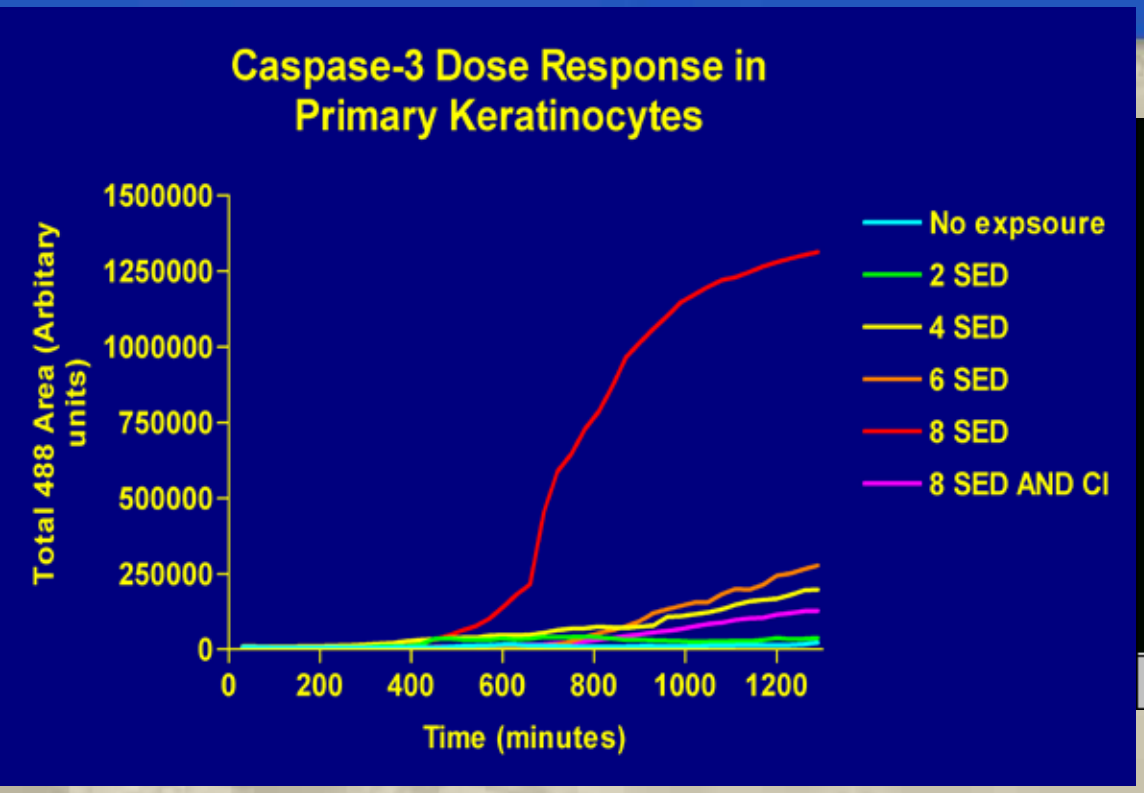
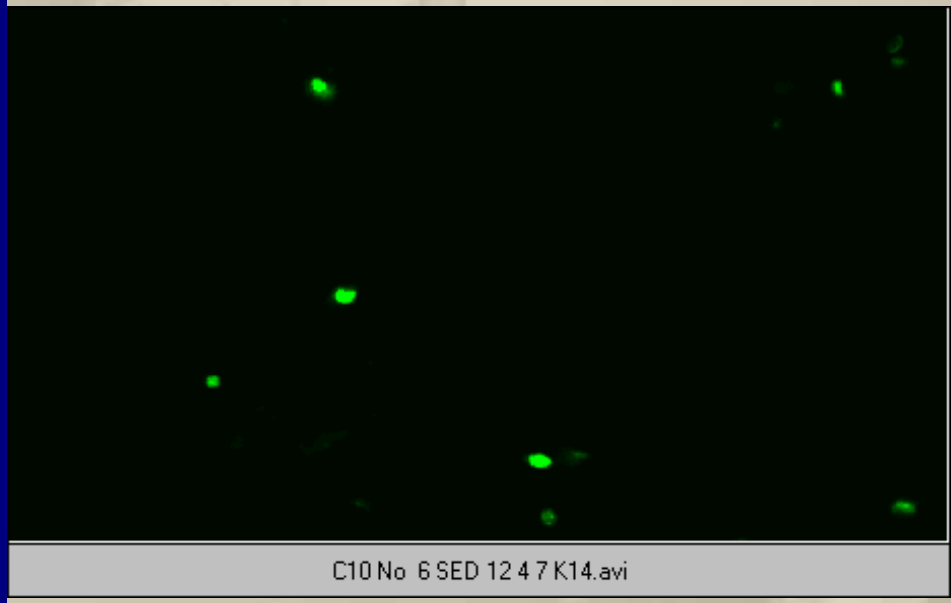
Apoptosis

Methods – NucView™ 488 Caspase substrate

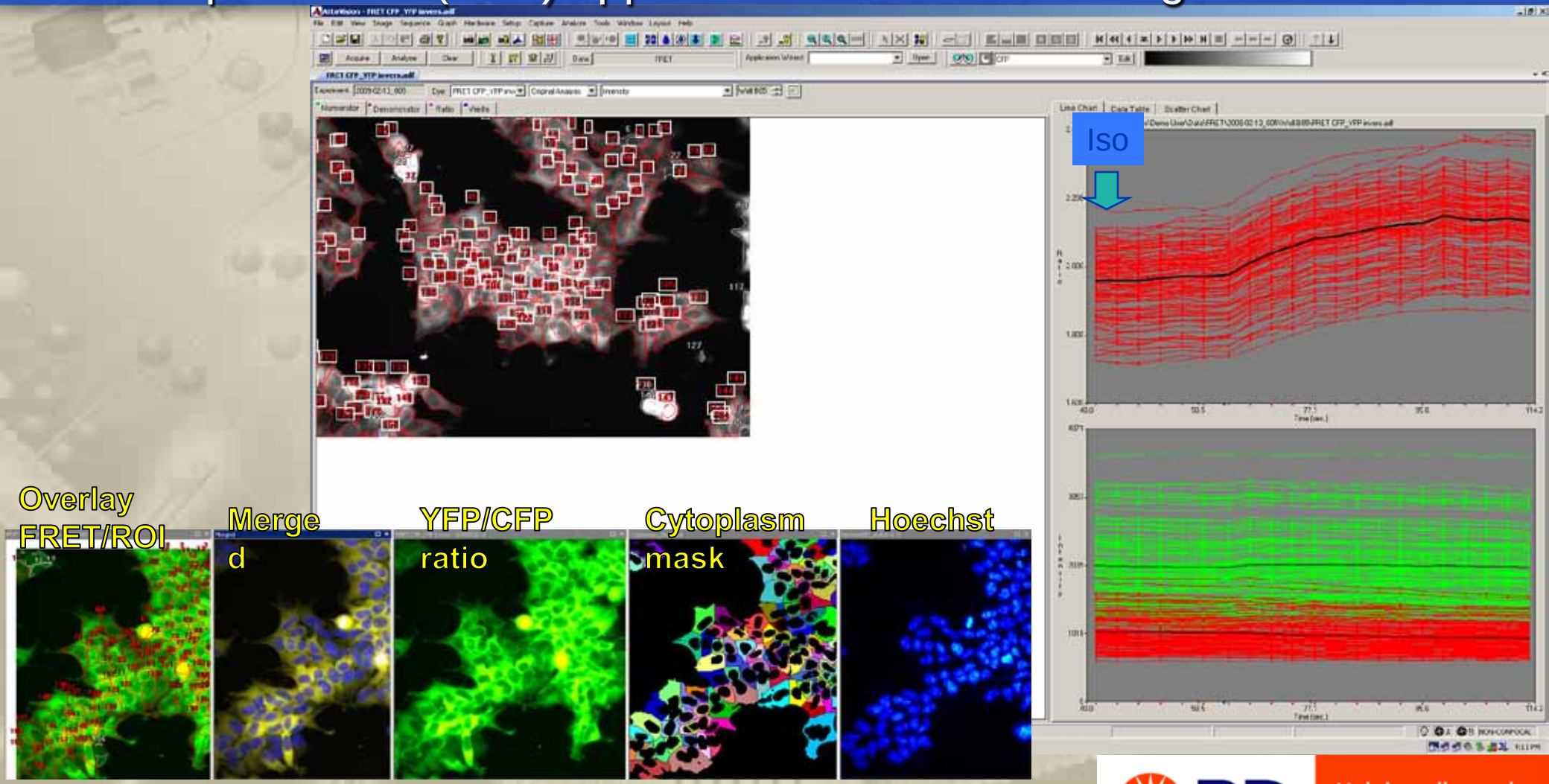


Effect of UV radiation on Caspase-3 activity





BD Pathway 855: Examples for Applications
FRET: Images and Segmentation of CFP/YFP FRET pair
Isoprotenerol (2uM) application results in a change of FRET



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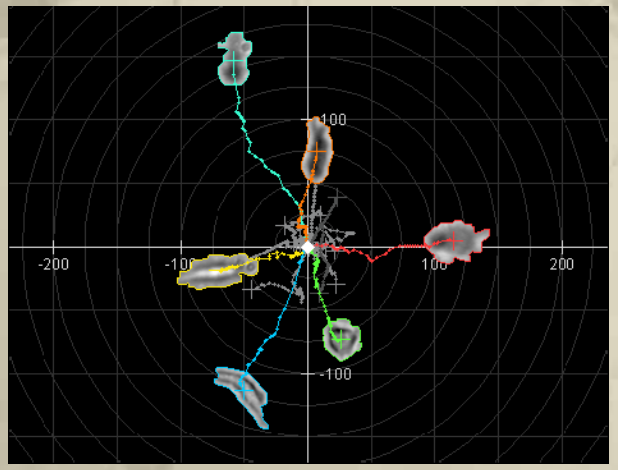
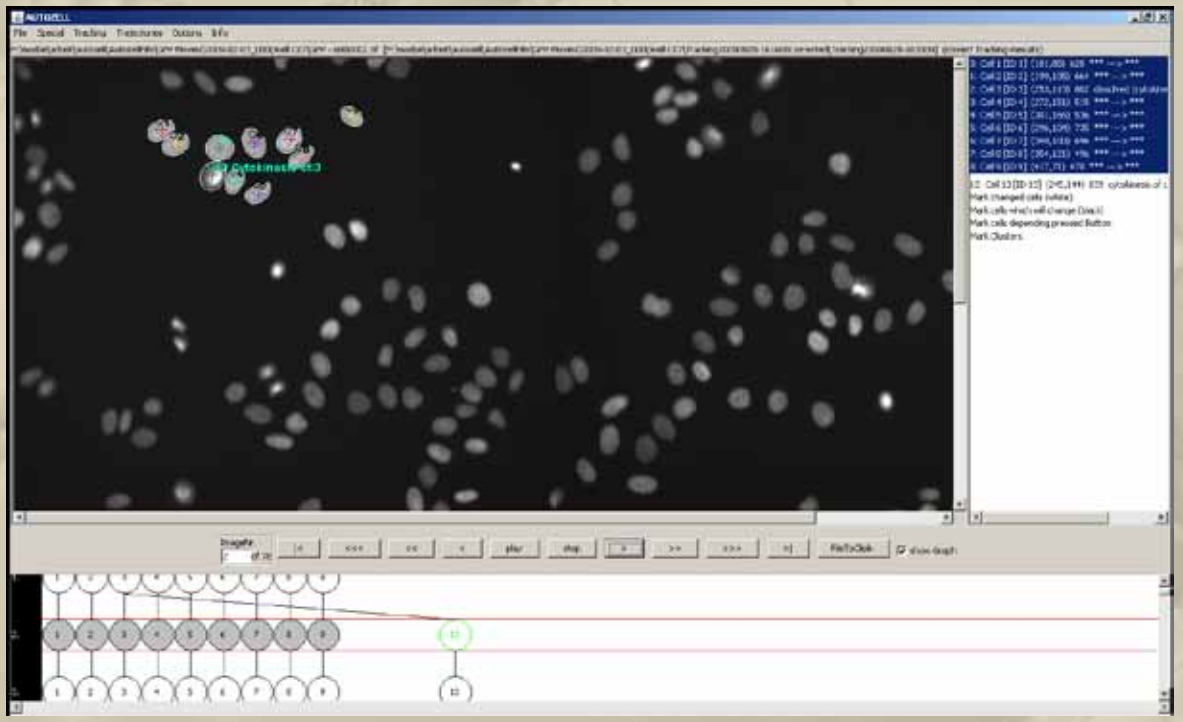
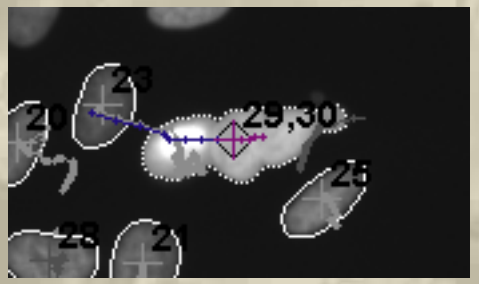
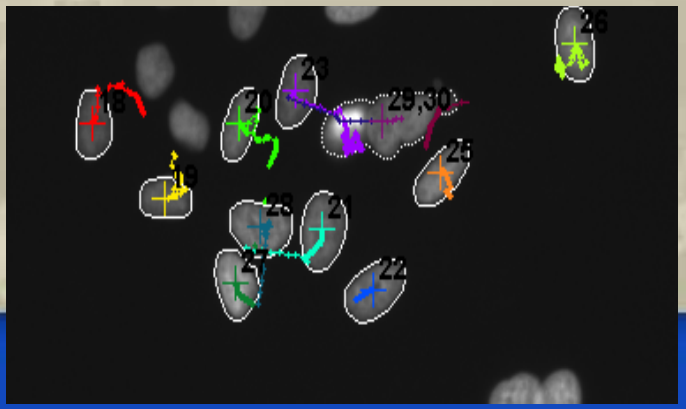
BD collaborates with TZI BREMEN to enable direct tracking analysis of Attovision time series with AUTOZELL tracking of cell division









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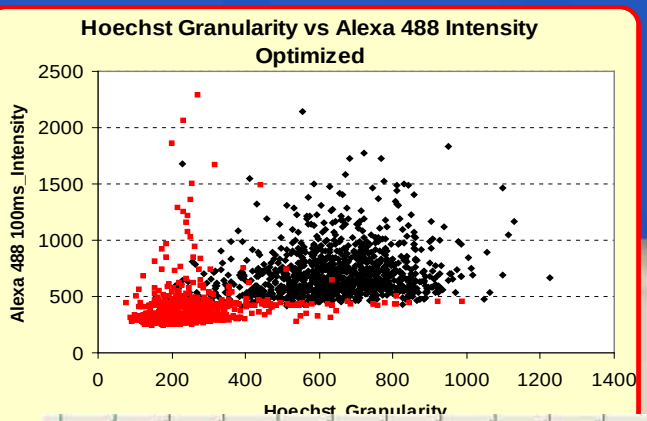
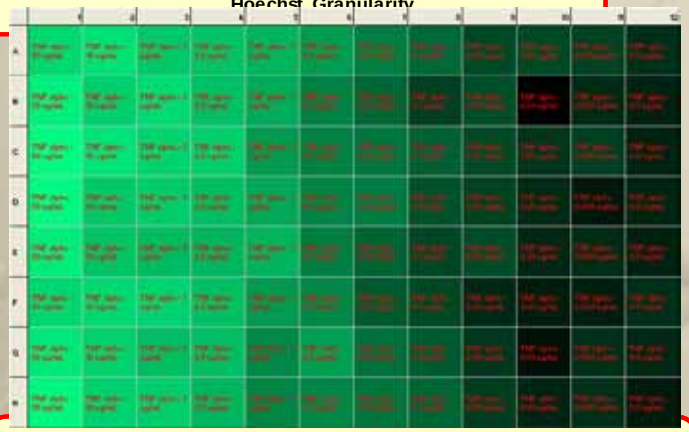
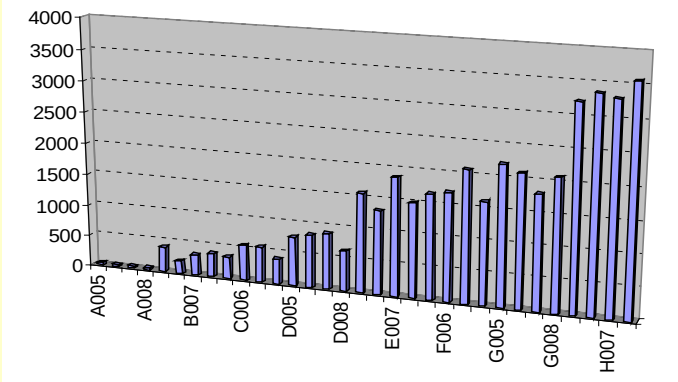
AttoVision Data Analysis Outputs

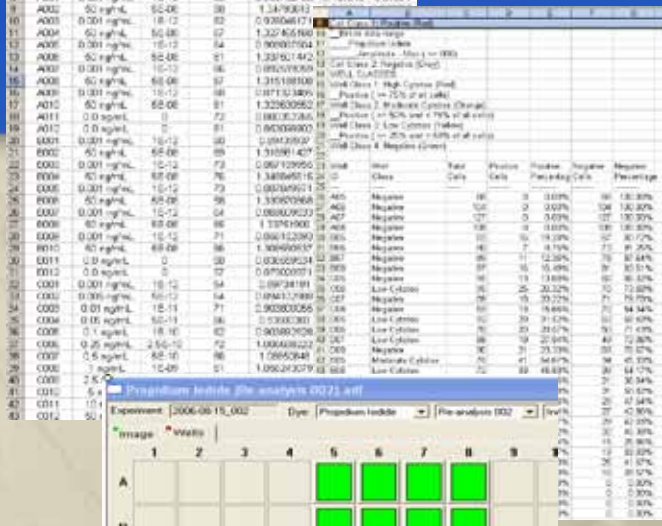
- Built in flexibility
  - Open architecture
- Choice of data output
  - Kinetic analysis
  - Data classification
  - EC/IC$_{50}$ curves
  - Bar charts
  - Scatter plots
  - Heat maps
  - Reports

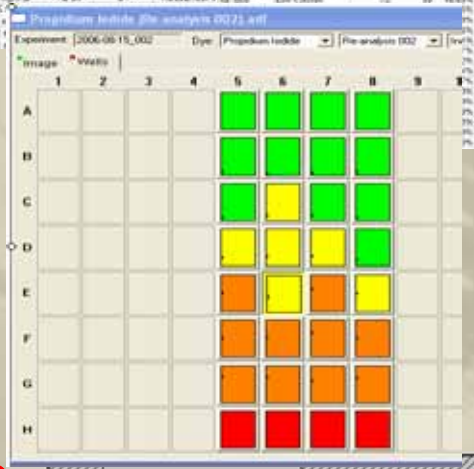


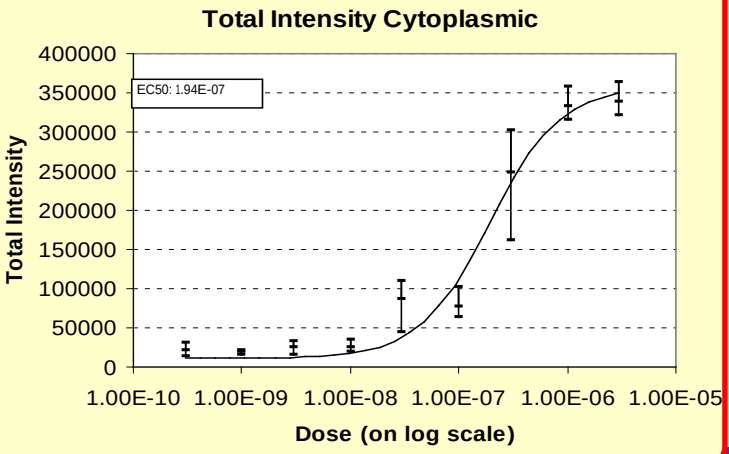


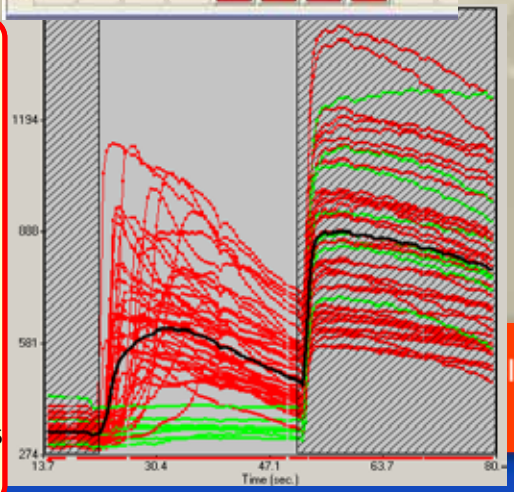
Average Intensity of PI





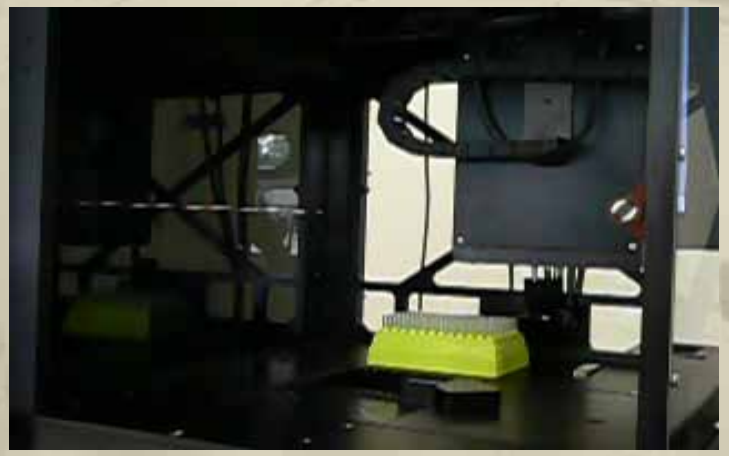


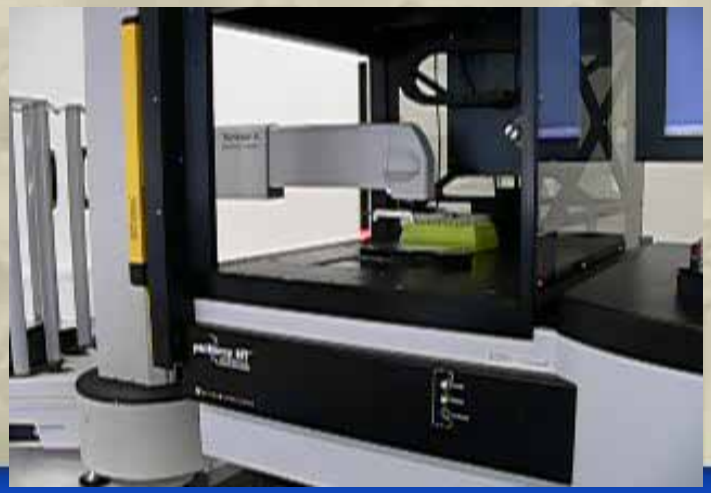




Integration with robotics:

BD Pathway Bioimager 855 + Caliper TWISTER II





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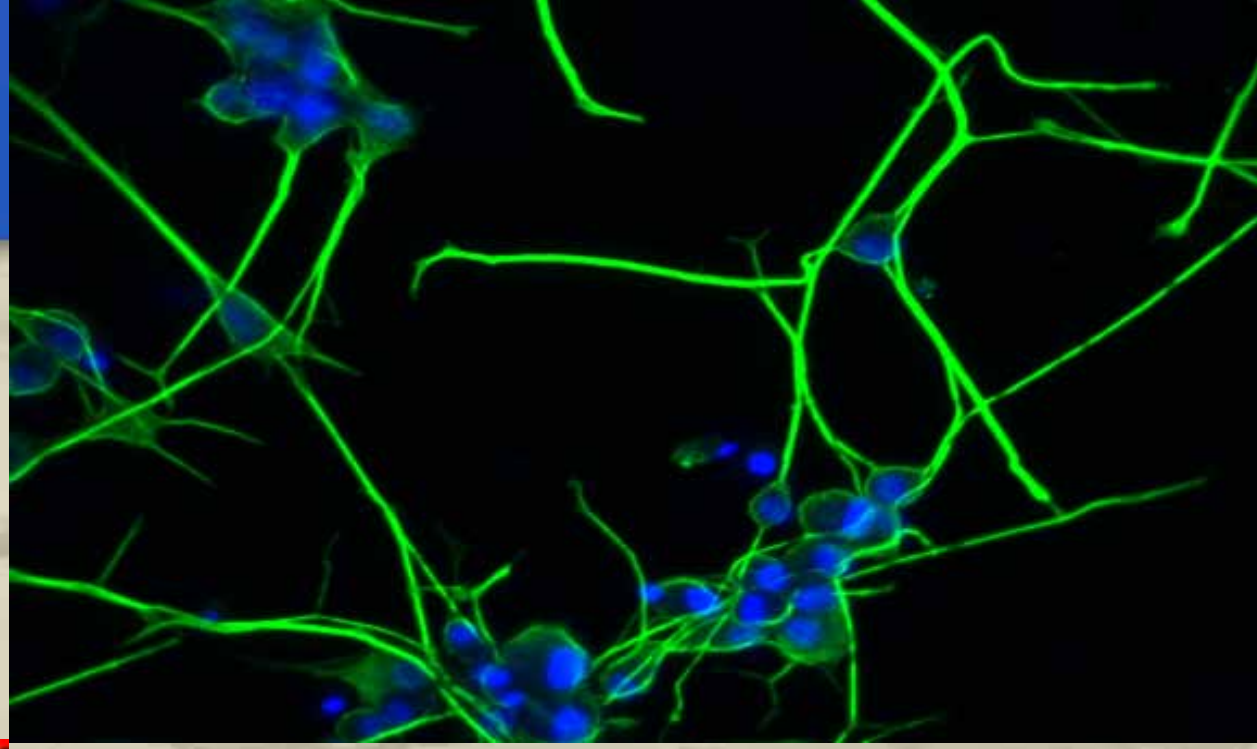
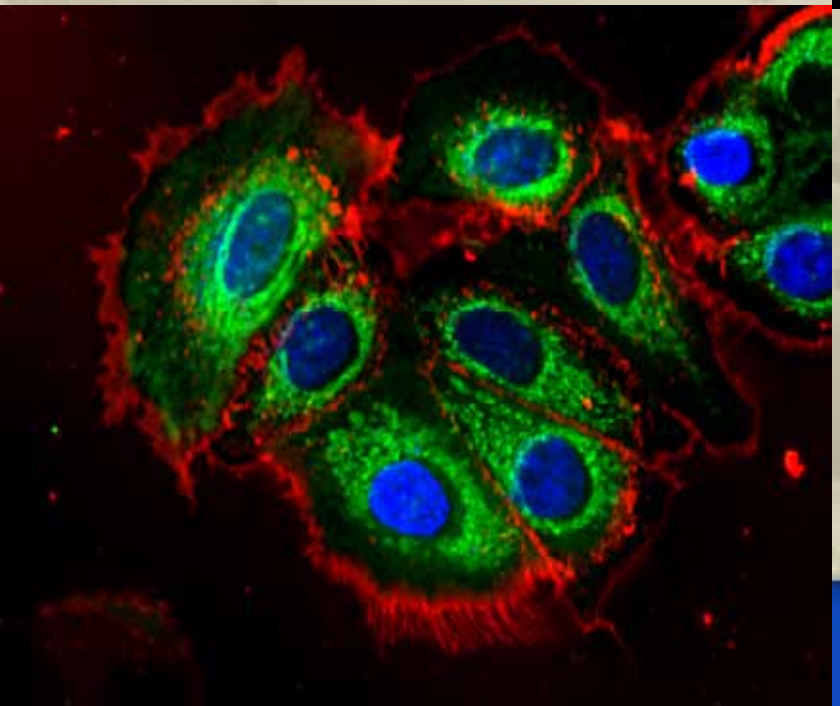
NANOSCIENCE 2009
Lichtenwalde

Thomas Horn, PhD
BD Biosciences
European Technology Center
Allschwil, Switzerland





Thank you!

Questions?

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