

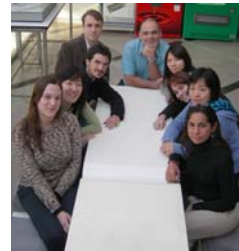
"High Content and High Throughput Screening applied to drug discovery for parasitic diseases"



•Strengths: Using phenotypic assays for drug discovery

Neglected diseases at Institut Pasteur Korea

Systems Biology of Pathogens



Malaria

Cases: 273 million infected annually
Deaths: 1.09 million annually
People at risk: 2.1 billion



Leishmaniasis

Cases: 12 million infected annually
Deaths: 57,000 annually
People at risk: 350 million



Chagas disease

Cases: 16-18 million infected
Deaths: 21,000 annually
People at risk: 120 million



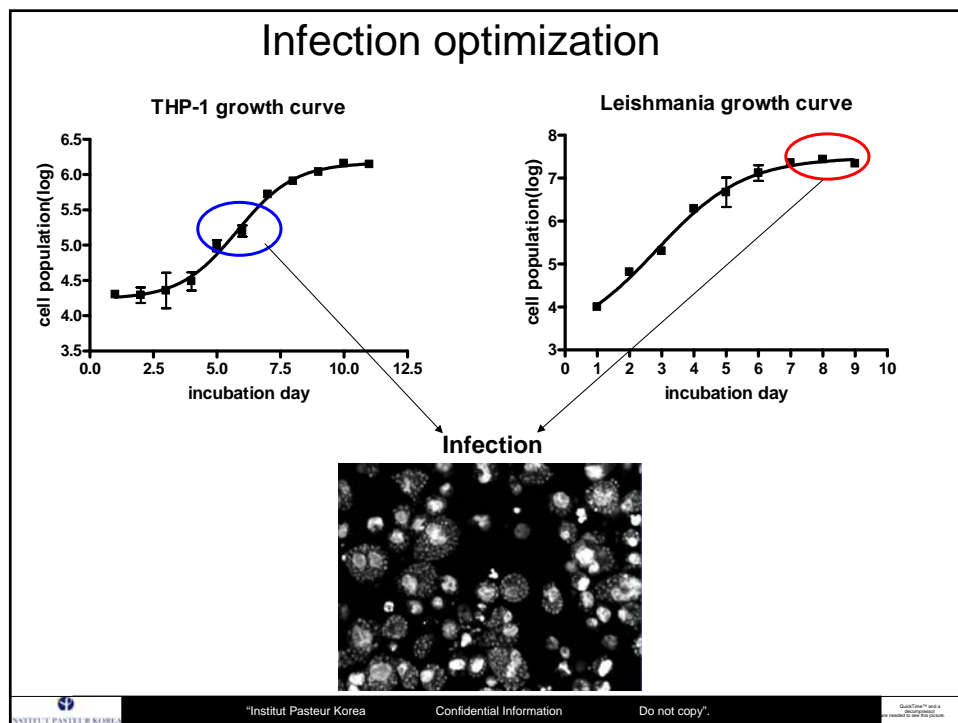
Integrated approach

INFECTIOUS DISEASE

IMAGE ANALYSIS

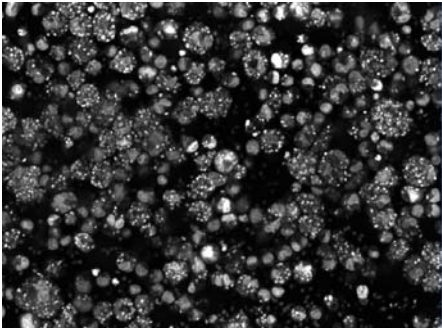
VISUAL SCREENING

Reading in Opera (Imaging)



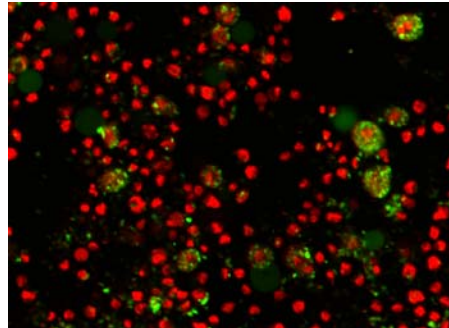
GFP assay problem

Leishmania WT



76% infection

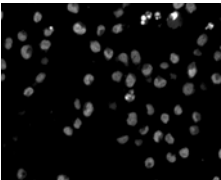
Leishmania GFP

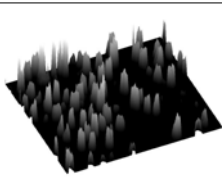


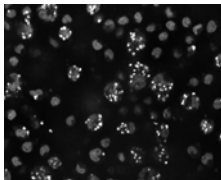
Angela Cruz
Universidade de
Sao Paulo

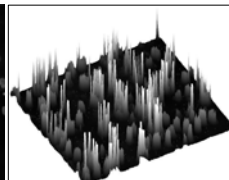
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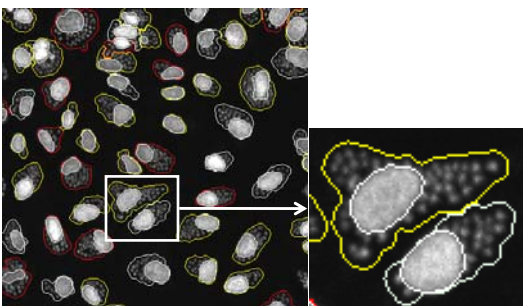
Need for an automated multidimensional analysis of macrophages infection







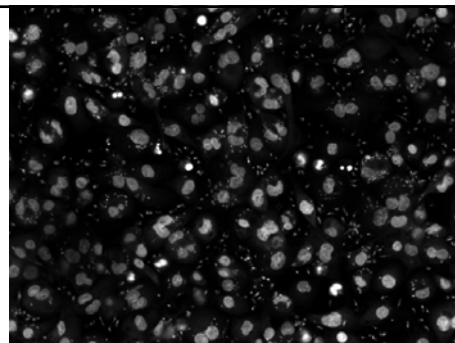
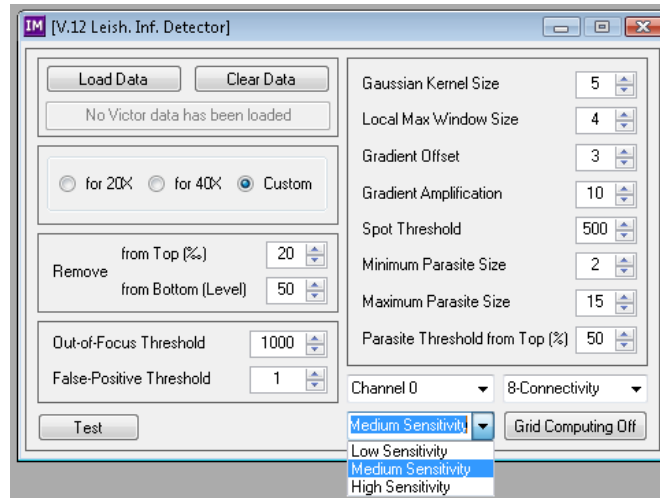




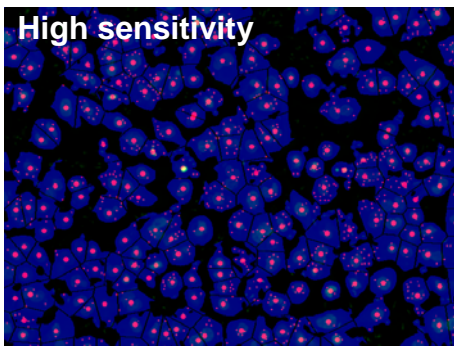
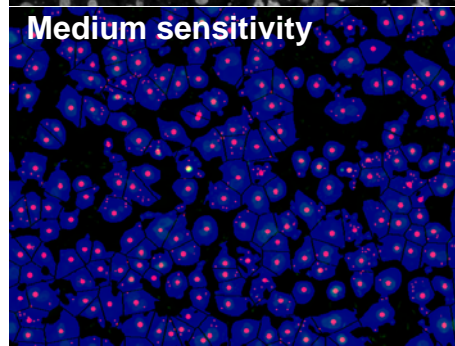
- cell number
- infected cell number
- number of parasites per cell
- cell dispersion
- cell shape/texture description

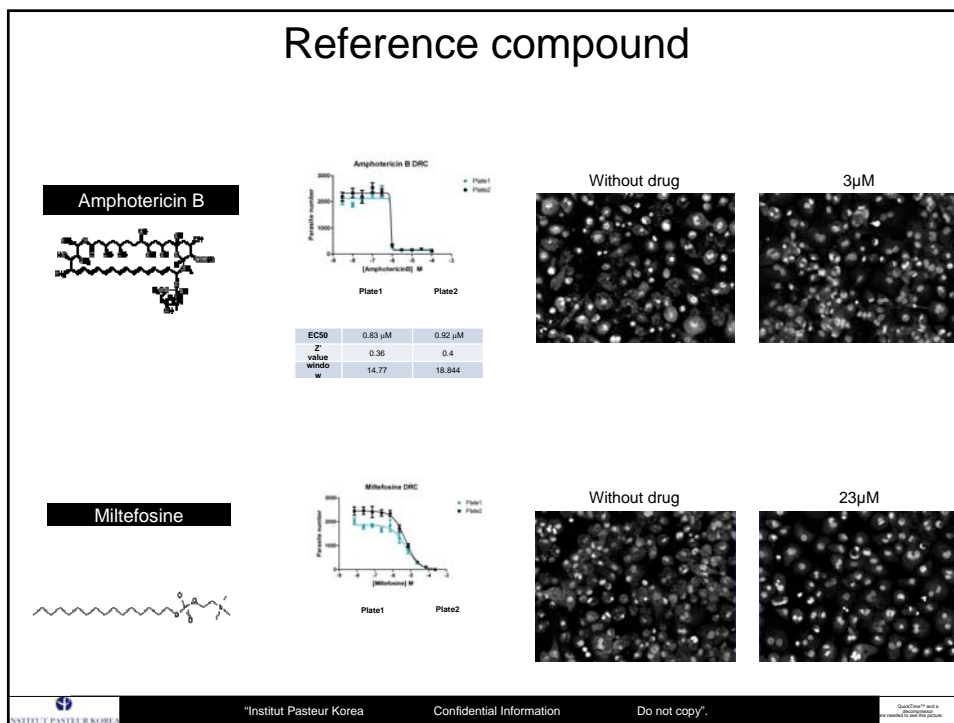
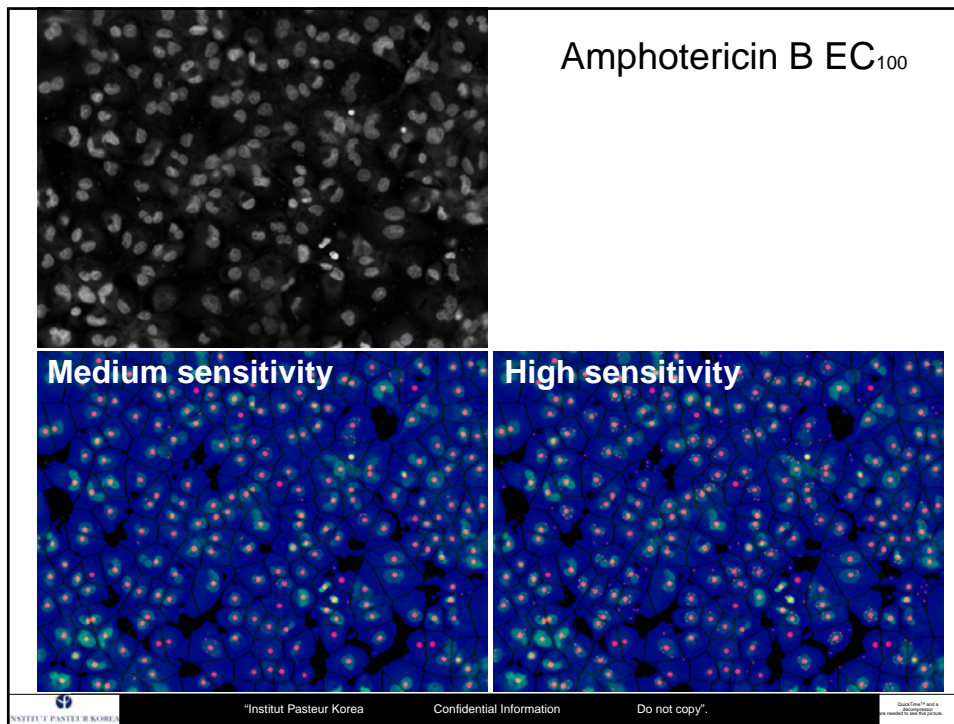
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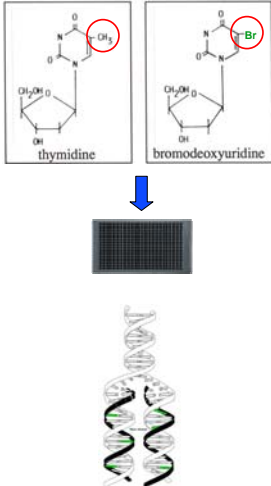
Dedicated algorithm...Analysis software



DMSO 1%









thymidine

bromodeoxyuridine

QuickTime™ and a decompressor are needed to see this picture.

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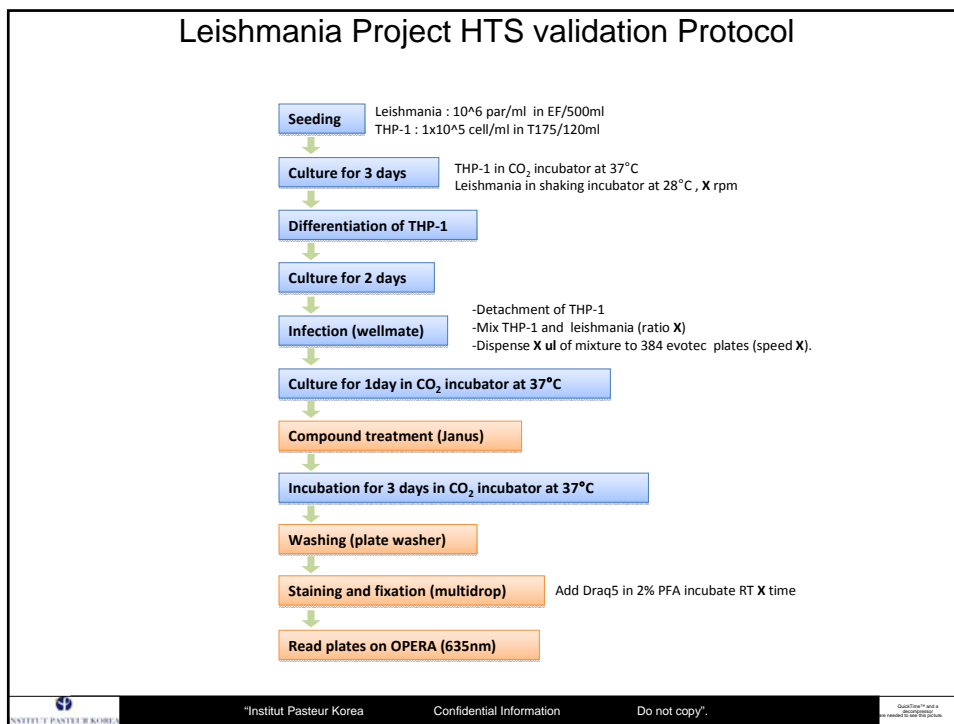
Technology



- IPK (Seoul)
- BSL-3 (HIV, TB)
 - 24 hour, unattended operation



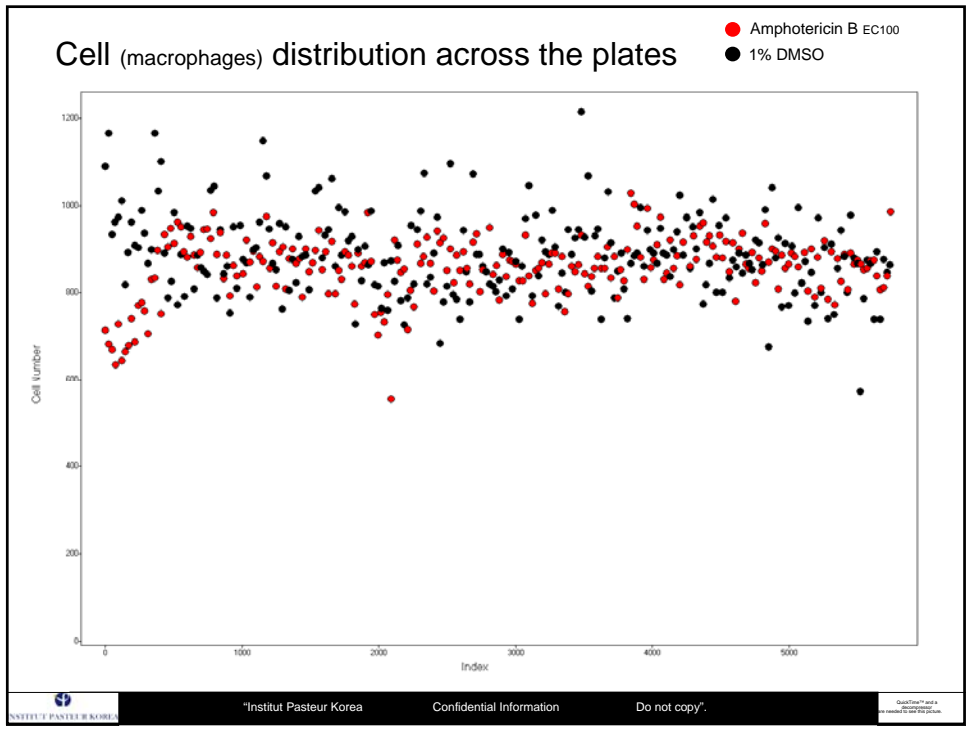
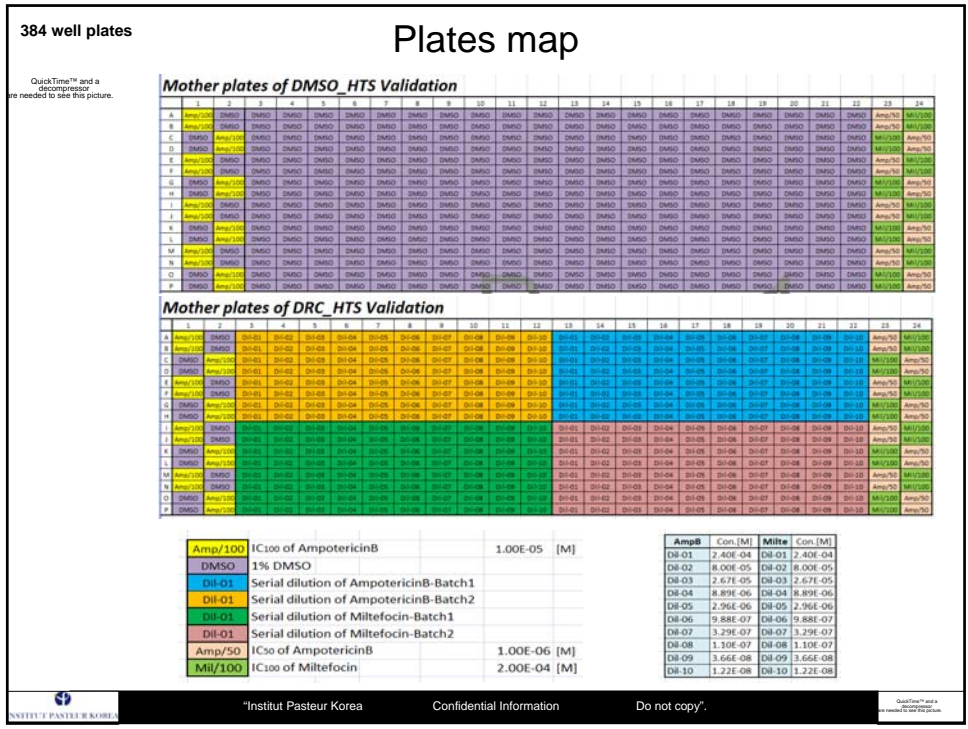
- IPK-GGBC (Suwon)
- BSL-2 (*Leishmania*, *T. cruzi*)
 - 24 hour, unattended operation

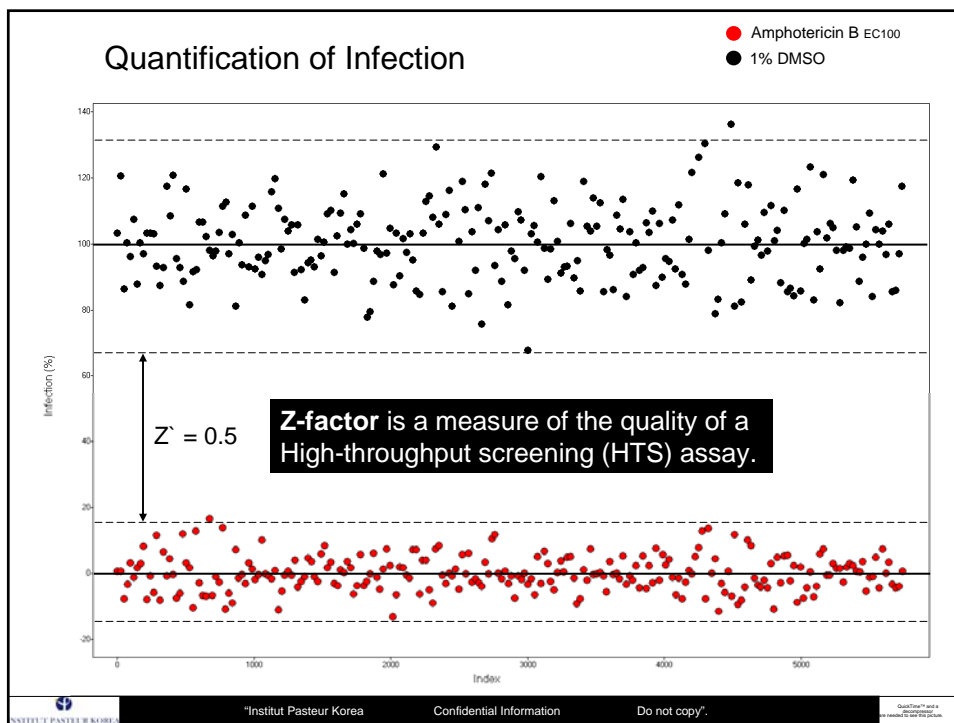
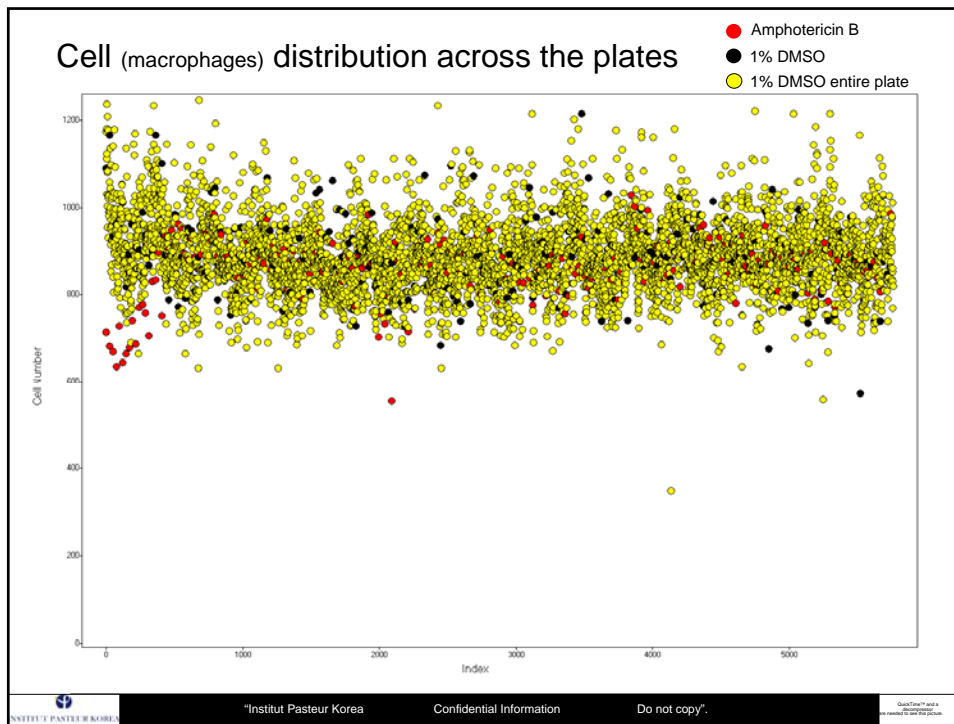


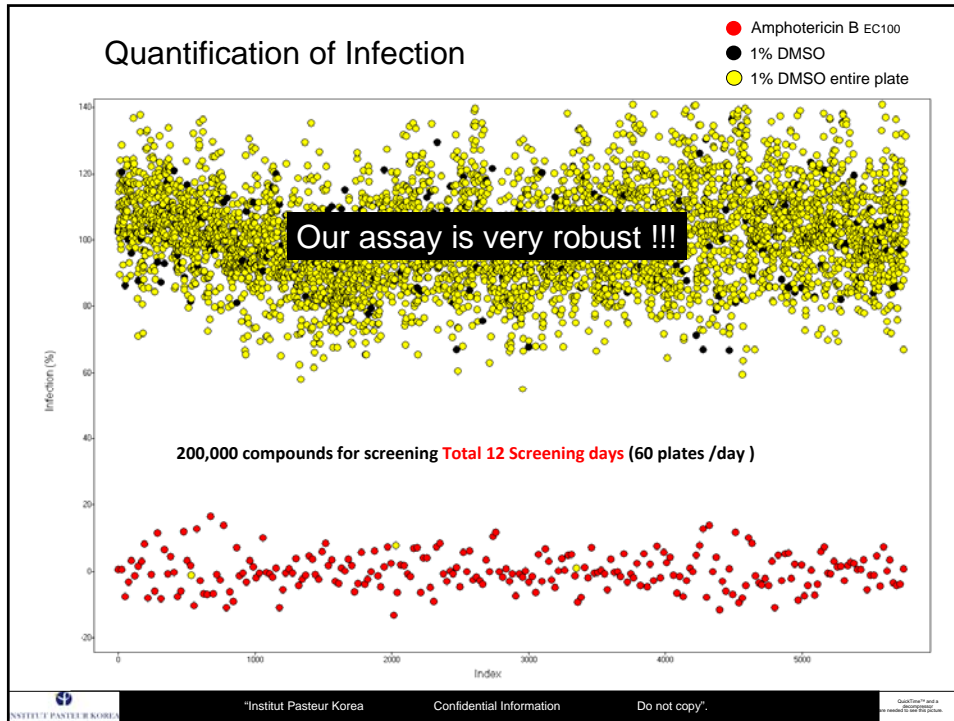
Day differences in the protocol

	HTS_V1	HTS_V2	HTS_V3	HTS_V4
Number of plates	40 plates	30 plates	30 plates	30 plates
Mixture preparation time	2hrs 30min	1hrs 20 min	1hrs	1hrs
Compound treatment by Janus	Tip washing with autoclaved DW	w/o tip washing	Tip washing with 3°DW	Tip washing with autoclaved DW
Drug con.	AmpEC100=1.00E-05 [M] AmpEC50=1.00E-06 [M] MilteEC100=2.00E-04 [M] in PBS	AmpEC100=1.00E-05 [M] AmpEC50=1.00E-06 [M] MilteEC100=2.60E-05 [M] in DMSO 1%	AmpEC100=1.00E-05 [M] AmpEC50=1.00E-06 [M] MilteEC100=2.60E-05 [M] in DMSO 1%	AmpEC100=1.00E-05 [M] AmpEC50=1.00E-06 [M] MilteEC100=2.60E-05 [M] in DMSO 1%
Contamination	NO	NO	YES in some wells	NO

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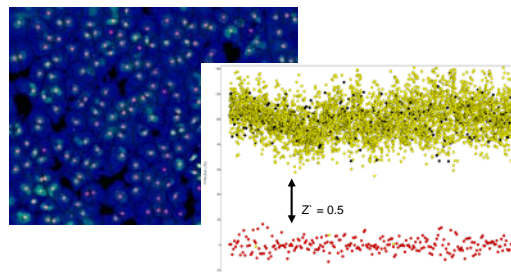


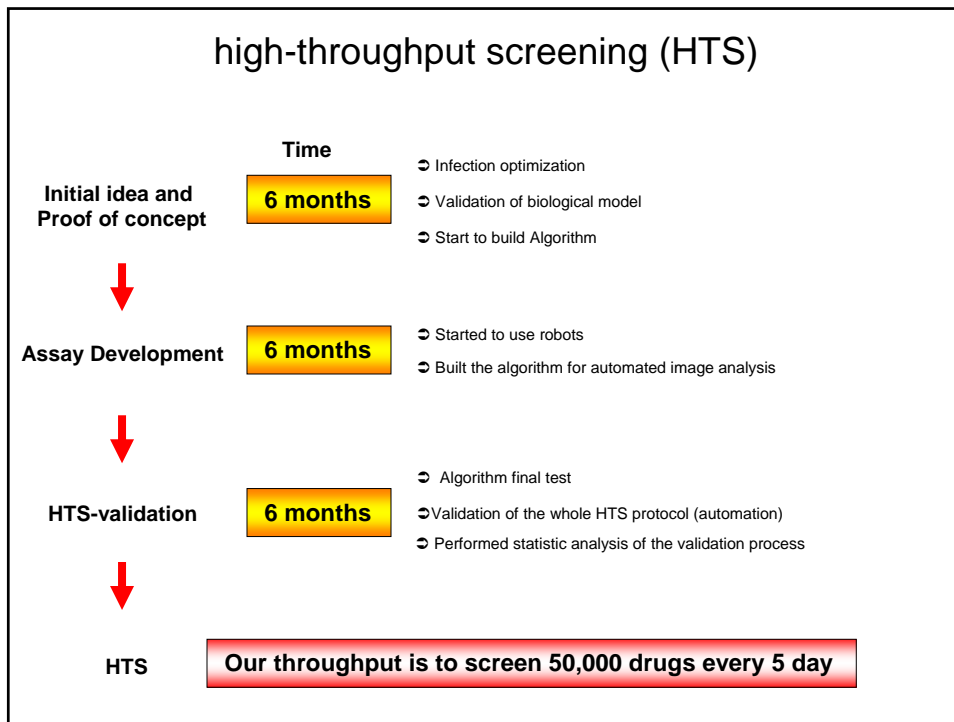
summary and conclusion


- Screening with the most appropriate and relevant cellular model of Leishmaniasis:

- Human macrophage cell line (THP1)
- *L. donovani*


- Robust assay:










INSTITUT PASTEUR KOREA



Acknowledgements

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Lucio Freitas Group Leader	Auguste Genovesio Group Leader	Thierry Christophe Group Leader
		

Systems Biology of Pathogens

Image mining

Screening Technology and Pharmacology

Jair Postdoc	Gyong Seon PHD Student	Seunghyun PHD Student	Arnaud Ogier Scientist	Boris Lenseigne Scientist	Jiyeon Jang Researcher	Jonathan Cechetto Scientist	ChangBok Lee Researcher
