



Analysis Sequence "PreScan_Analysis XYZ Gri3D"

Input Image	Input		
	Channel group : 1 Sequences : ALL Flatfield Correction : None Brightfield Correction Stack Processing : Maximum Projection Create Global Image Min. Global Binning : Dynamic		
Filter Image	Input	Method	Output
	Channel : SPY555-DNA (global)	Method : Sliding Parabola Curvature : <u>8</u>	Output Image : Sliding Parabola (global)
Find Image Region	Input	Method	Output
	Channel : Sliding Parabola (global) ROI : Imaged Area (global) ROI Region : Imaged Area	Method : Whole Image Region	Output Population : Whole Image (global) Output Region : Whole Image Region
Select Region	Input	Method	Output
	Population : Whole Image (global) Region : Whole Image Region	Method : Resize Region [%] Outer Border : <u>5</u> % Outer Population : None Outer Region : Inner Border : 100 %	Output Region : Resized Region
Find Image Region (2)	Input	Method	Output
	Channel : Sliding Parabola (global) ROI : Whole Image (global) ROI Region : Resized Region	Method : Absolute Threshold Lowest Intensity : \geq <u>300</u> Highest Intensity : \leq INF Split into Objects Area : $>$ <u>300</u> μm^2 Fill Holes	Output Population : ROI Candidate Output Region : ROI
Modify Population	Input	Method	Output
	Population : ROI Candidate Region : Organoid	Method : Cluster by Distance Distance : <u>2</u> px Area : $>$ 0 px ²	Output Population : ROI Candidate Clustered Output Region : ROI Candidate Clustered Region

Calculate Intensity Properties	Input	Method	Output
	Channel : SPY555-DNA (global) Population : ROI Candidate Clustered Region : ROI Candidate Clustered Region	Method : Standard Mean	Property Prefix : ROI Candidate Clustered Intensity SPY555
Calculate Morphology Properties	Input	Method	Output
	Population : ROI Candidate Clustered Region : ROI Candidate Clustered Region	Method : Standard (deprecated) Area Roundness Width	Property Prefix : ROI Clustered Morphology
Select Population	Input	Method	Output
	Population : ROI Candidate Clustered	Method : Filter by Property ROI Clustered Morphology Roundness : > <u>0.2</u> ROI Clustered Morphology Area [μm^2] : > <u>2000</u> ROI Clustered Morphology Area [μm^2] : < <u>100000</u> Boolean Operations : F1 and F2 and F3	Output Population : ROI Selected
Determine Well Layout 	Input	Method	Output
	Population : ROI Selected Region : ROI Candidate Clustered Region	Method XY : Individual Objects Rescan Magnification : 20x Rescan Camera ROI : 2160x2160 Max No of Fields : Object Margin : <u>20</u> μm Field Overlap : 2 % Method Z : By Plane Map Plane Map : Plane Map SPY555-DNA (global)	Output Population : Well Layout (global)
Define Results 	Results		
	Method : Standard Output Clustered Organoid Candidate - Number of Objects : Object Count Output Name : Clustered Organoid Candidate - Number of Objects Method : List of Outputs		

Object Results

Population : Whole Image (global) : None
Population : Clustered Organoid Candidate : None
Population : Organoid Candidate : None
Population : Clustered Organoid Candidate Selected : None

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