IKOSA®
Application Documentation

Application Name	Spheroid Sprouting Assay
Version	1.0.0
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Input Image(s)	Single Image (standard, grayscale) RGB images are automatically converted to grayscale images
Input Parameter(s)	None
Keywords	Spheroid, sprouting, sprout, formation, in-vitro, angiogenesis, vessel, growth, microscopy
Short Description	Detection and quantification of sprouts in a spheroid capillary sprouting assay used for in-vitro angiogenesis research.
References / Literature	For more information regarding the assay check e.g. https://www.ncbi.nlm.nih.gov/pubmed/23911327; Reference laboratory: Department of Obstetrics and Gynecology: Dr. Ursula Hiden; Jasmin Strutz, MSc;

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IKOSA® Image Analysis

You can use this or any other of our image analysis applications through your IKOSA® account. If it is not listed in the available applications, please contact your organization’s IKOSA® administrator or our team at support@kmlvision.com.

Application Description

This application automatically segments the body and sprouts created by endothelial cell spheroids in a 3D collagen matrix, and extracts relevant measures (number of sprouts, sprouting lengths, body measures).

In the following, the requirements for an accurate analysis are given and the output of the applications is described.

Further Information

If you have any questions regarding this application or if you want to know if your specific type of images can be analyzed, please get in touch with us at support@kmlvision.com. Also, if you have requests or ideas regarding additional image analysis applications that you would require, please get in touch with us at support@kmlvision.com.

For more information, please visit www.ikosa.ai.

Requirements

Input Image(s)

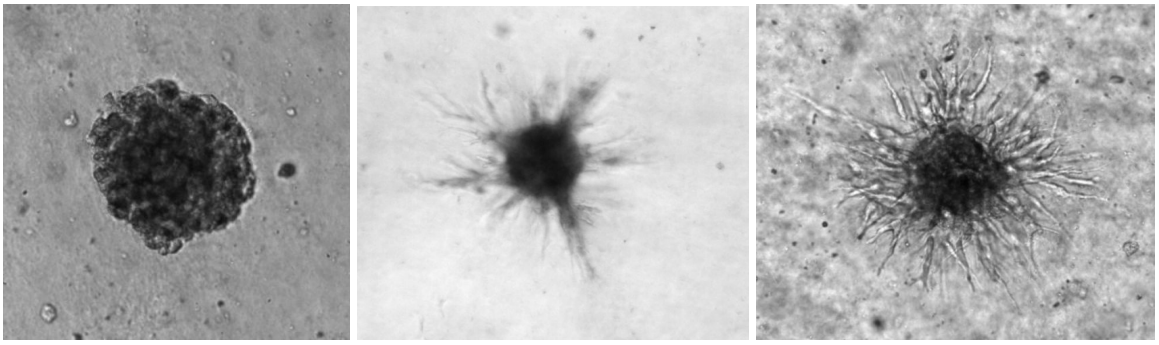
Input for this application is the following image data:

No.	Image data	Type of image	Color Channels	Color Depth (per channel)	Size [Px]	Resolution [µm/Px]
#1	Single image	Standard	1 (Greyscale) 3 (RGB)	8 Bit	Min: 1280 x 1280 Max: 4096 x 4096	typically: 0.5 - 1.0

Image Content: Transmitted light microscopy image of spheroid capillary sprouting assay, typically taken with 10x magnification. In case of multiple bodies in the image, the body closest to the image center is detected and used for sprout analysis.

Additional requirements: None

Examples:



For all images, the following requirements apply:

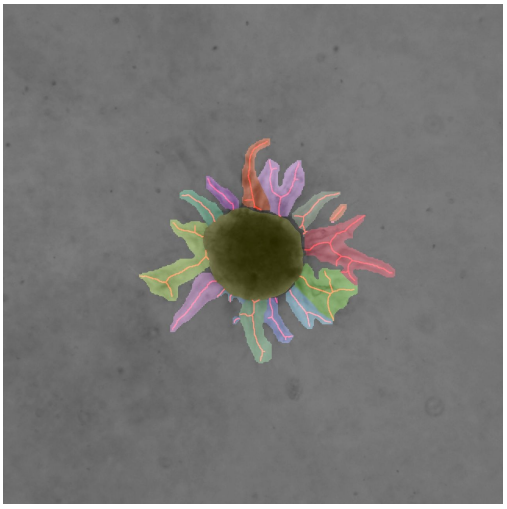
- The illumination must be constant throughout the image(s).
- The sample must be in focus, i.e. no blurry regions in image(s).

Input Parameter(s)

No additional input parameters are required for this application.

Results

Files

No.	File type	Content and Description
1	csv	*results.csv:* A csv file containing the analysis results for the input image.
2	jpg	*results_visualization.jpg*: A visualization of the detection:  • The segmented body is shown in yellow, while different sprouts are shown in different other colors. • In case of multiple bodies in the image, the body closest to the image center is detected and used for sprout analysis.. • The skeleton, which is used to calculate the sprouting length, is shown in red.

Description of files

File no. 1: Single csv-file with the following content (*results.csv*):

Col. no.	Column name	Examples	Value range	Description
1	total length sprouts [Px]	104	0 -	Total length of all sprouts in pixels.
2	number of sprouts	26	0 -	Number of detected sprouts.
3	body area [Px^2]	58775	0 -	Area of body in pixels^2.
4	body circularity	0.7644	0 - 1	Circularity of the body calculated as $f_{\text{circ}} = \frac{4\pi A}{P^2}$ where **A** denotes the area of the body and **P** the perimeter of the body. **Note:** Due to the discrete nature of images, the calculation of the perimeter can only be approximated. Therefore, the circularity of a body being a circle, results in a value of approximately 0.9. Theoretically, a circle has a value of 1.0. [1] [1] Bottema, Murk J. "Circularity of objects in images." *2000 IEEE International Conference on Acoustics, Speech, and Signal Processing. Proceedings (Cat. No. 00CH37100)*. Vol. 4. IEEE, 2000.