

# In vitro endothelial spheroid sprouting assay for angiogenesis

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## Contact person

Ward De Spiegelaere

## Organisation

**Name of the organisation** Ghent University (UGent)

**Department** Morphology

**Country** Belgium

**Geographical Area** Flemish Region

## SCOPE OF THE METHOD

<b>The Method relates to</b>	Animal health, Human health
<b>The Method is situated in</b>	Basic Research
<b>Type of method</b>	In vitro - Ex vivo
<b>Species from which cells/tissues/organs are derived</b>	Human
<b>Type of cells/tissues/organs</b>	endotelial

## DESCRIPTION

### Method keywords

collagen

endothelial

sprouting  
spheroids

### **Scientific area keywords**

angiogenesis  
tip cell  
endothelial sprouts  
sprouting angiogenesis

### **Method description**

This method evaluates sprouting propensity of endothelial cells in a collagen matrix. The effect of pro-angiogenic growth factors or co-cultured cells can be measured by quantifying the amount of vascular sprouts that form on endothelial spheroids. Endothelial spheroids are obtained by growing endothelial cells in hanging drops, which forces the cells to adhere to each other. The peripheral cells acquire a flat phenotype, similar to the flattened endothelial wall of blood vessels. After generation of the spheroids, they are embedded in a collagen matrix in which endothelial growth factors or specific cell types can be embedded. Finally the amount of endothelial sprouts is quantified as a measure of the endothelial sprouting propensity.

### **Lab equipment**

Biosafety cabinet ;  
CO2 incubator ;  
Inverted microscope.

### **Method status**

Internally validated

## **PROS, CONS & FUTURE POTENTIAL**

### **Advantages**

The method is simple and the vascular sprouts share multiple morphological characteristics of vascular tip cells *in vivo*.

## Challenges

The model is limited to evaluating sprout propensity, which is only the first step in the angiogenic cascade. The subsequent steps of tubule and network formation cannot be evaluated.

## Modifications

Spheroids can also be made from a mix of cells. We have performed this with hepatic cancer cells and this lead to a core of cancer cells surrounded by a flattened endothelial layer, similar to an inside out blood vessel.

## REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

### References

Ffisterer, L. Korff, T. (2016) Spheroid-Based In Vitro Angiogenesis model. Methods Mol Biol; 1430:167-77 doi: 10.1007/978-1-4939-3628-1\_11

### Links

[An alternative spheroid sprouting protocol](#)

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