

In vitro endothelial spheroid sprouting assay for angiogenesis

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Organisation

Name of the organisation Ghent University (UGent)

Department Morphology

Country Belgium

Geographical Area Flemish Region

SCOPE OF THE METHOD

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

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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