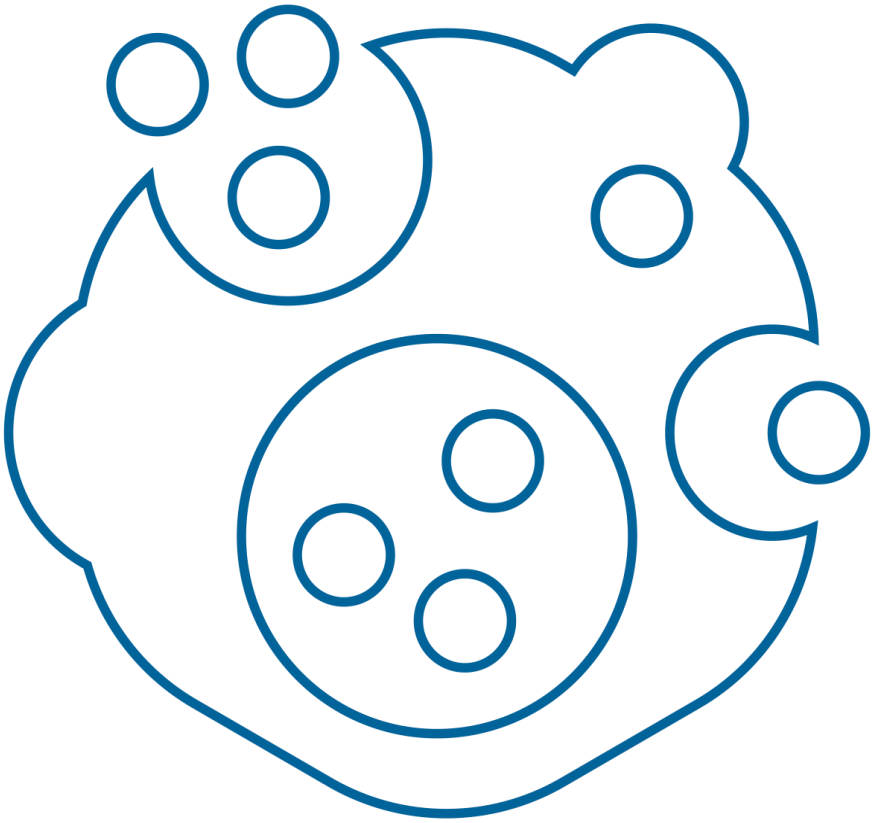
Forever Is Just Enough

EVERCYTE



EV toolbox

From production to characterization

Good experiments start with the right choices – hTERT immortalized cell lines retain the cell-type specific phenotype while constantly growing.
No more lot-to-lot variability.
No more growth arrest.
Just the perfect choice!

Extracellular vesicles– in a nutshell

EV cell factories: telomerized xeno-free MSCs from adipose tissue, bone marrow, dental pulp, endometrium, amnion, placenta and Wharton's jelly

EV characteristics: presence of CD9, CD81, CD63, typical EV size and double-layer membrane

Cell specific *in vitro* biological activity (anti-inflammatory, anti-fibrotic, neo-angiogenic and wound, cartilage-healing)

EVs towards clinical application
EVs produced by human MSC cell lines established under xeno-free conditions with full documentation!

Extracellular vesicles from human telomerized cells

Extracellular vesicles (EVs) play an essential role in cellular communication by transporting proteins, lipids as well as nucleic acids. Thus, EVs have attracted the attention of biomedical research in immunotherapy, anti-tumor therapy, or regenerative and transplant medicine, as EVs secreted from e.g. human mesenchymal stromal cells have been shown to be equally effective as the transplanted cells in different studies. Thus, using EVs instead of cells might reduce regulatory burden and allow for therapeutic off-the-shelf products.

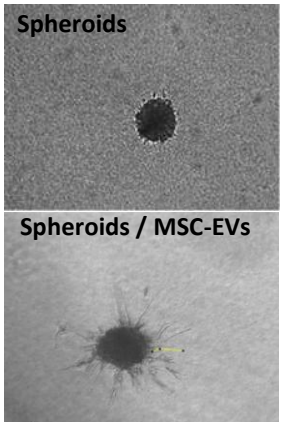
Currently, Evercyte EV cell factories portfolio consist of MSC from adipose tissue (ASC/TERT300), bone-marrow (BM-MSC/TERT292), dental pulp (DPSC/TERT349), endometrium (EN-MSC/TERT352-B), placenta (CP-MSC/TERT308), amnion (P-MSC/TERT308) and Wharton's jelly (WJ-MSC/TERT273).

Human production hosts

- Tissue sourcing and establishment of primary cells under xeno-free conditions with full documentation
- Lifespan extension of primary cells by ectopic expression of hTERT and/or cell cycle regulators using non-viral gene transfer
- Characterization of cells for expression of cell type specific markers and function, quality control testing, cell stability, identity, growth potential

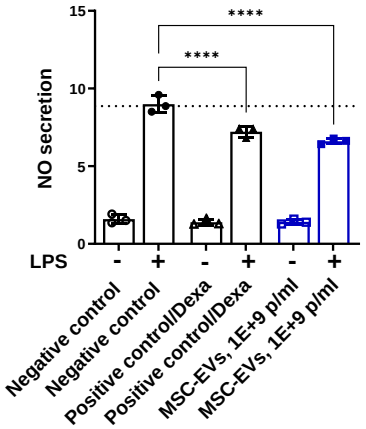
Extracellular vesicles

- Characterization of EVs for particles concentration/size (NTA), analysis of protein content and presence of EV markers (beads-based flow cytometry), morphology (cryo-EM)
- *In vitro* assays to test the EV biological activity (e.g. neo-angiog enic, anti-inflammatory, anti-fibrotic, wound-healing assay)
- Scale-up production and RNAseq/proteomics services by EV*scale*™ technology platform (partnership between Evercyte, Phoenestra and TAmiRNA)



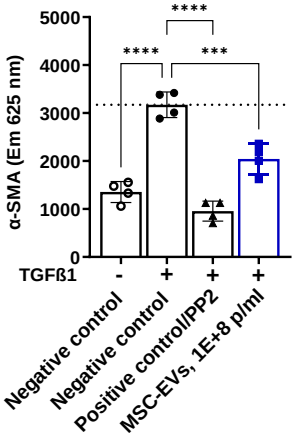
Neo-angiogenesis

Treatment of endothelial spheroids with MSC derived EVs induces the formation of sprouts, indicating a neo-angiogenic potential of such EVs.



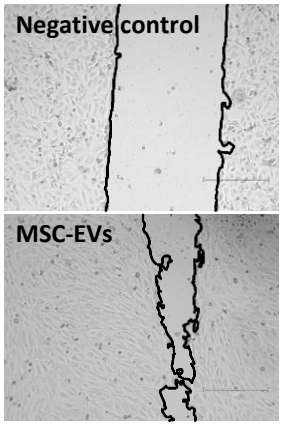
Anti-inflammation

Addition of MSC derived EVs to LPS induced macrophages significantly reduces NO secretion indicating an anti-inflammatory activity of the EVs.



Anti-fibrosis

Treatment of dermal fibroblasts with TGF-ß1 induces expression of α-SMA. MSC-derived EVs significantly reduce α-SMA induction, indicating anti-fibrotic activity.



Wound-healing

A physical gap within the monolayer of telomerised human endothelial cells is created and monitored over 40h. The addition of MSC-EVs, significantly promotes the gap closure.

Evercyte has focused on the establishment of human cell lines that allow standardizable production of high-quality extracellular vesicles

Evercyte is committed to follow the principles of Good Cell Culture Practice (GCCP, Pamies et al. 2022).

Our production host cell lines are:

- established following ethical standards (approved by IRB) with prior given written informed consent
- quality tested (sterility, absence of specific human-pathogenic viruses, STR-profile, longevity)
- characterized for presence of cell type specific markers and functions

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