Updated on: 31st March 2023

# CERTIFICATE OF ANALYSIS

**Lot#:** CHM2114-SC-P3-Z

## PRODUCT DESCRIPTION

**Reference:** HuSC
**Product:** Cryopreserved Human Stellate Cells
**Cellular Passage:** P3
**Size/Quantity:** > 100.000 cells/vial

**Isolation date:** 8th November 2021
**Storage conditions**: -196°C using $LN_2$
**Sterility test:** negative for mycoplasma, bacteria, yeast, and fungi

## DONOR DEMOGRAPHICS

| Species | Gender | Race | Age | BMI | Smoker | Alcohol Use | Drug Use |
|---|---|---|---|---|---|---|---|
| Human | Male | Caucasian | 72 | 22.49 | No | No | No |

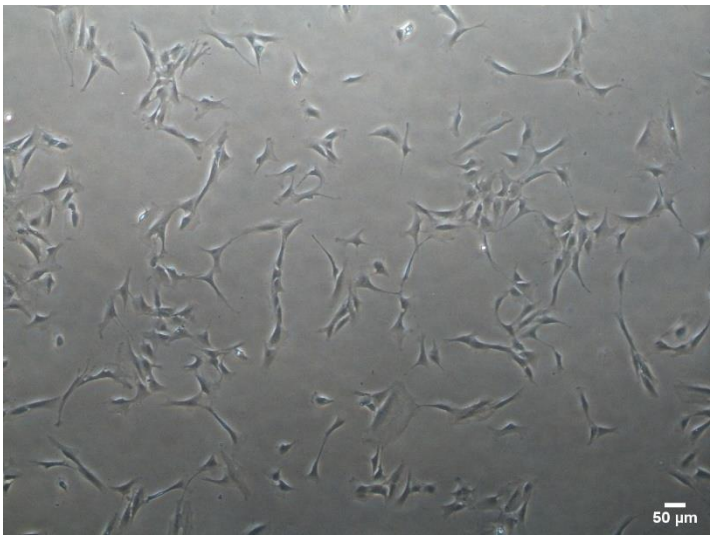
| Pathology | Serological Data[1] |
|---|---|
| Colorectal Cancer | Tested negative less than 3 months before surgery |

Patient informed consent was obtained. [1]The donor was serologically tested negative for following infectious diseases: HIV, Hepatitis B and C, and SARS-CoV-2. Donor medical history was also examined prior to accepting this donor. *For donor's medication information, please contact us.*

## CHARACTERIZATION FOR HUMAN STELLATE CELLS

| Post Thaw Lot information | |
|---|---|
| **Number of viable cells/vial:** | > 100.000 |
| **Cell seeding density (cells/cm²):** | 5.000-8.000 |

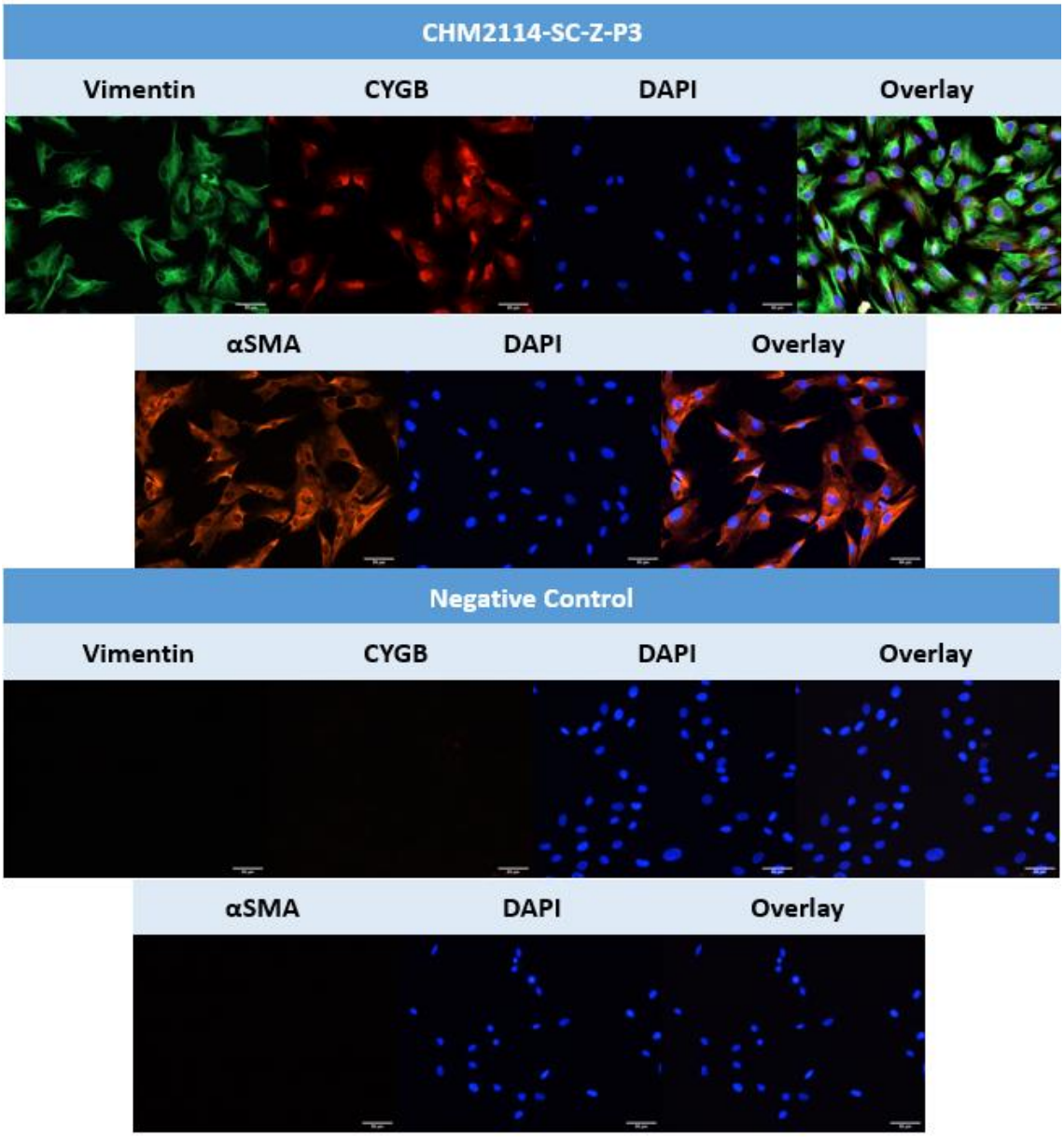
**Cell morphology**




Human stellate cells were thawed and seeded according to BeCytes Biotechnologies protocol. The number of cells and viability post-thawing was assessed by using the trypan blue exclusion assay. Phase-contrast image 5 days after seeding is shown on the panel.

## IMMUNOFLUORESCENCE ANALYSIS

Human stellate cells are positive for vimentin, and cygb and negative for α-smooth muscle actin when they are quiescent. When they become activated, they start to express α-smooth muscle actin.




Cells were cultured on 8 well chamber slide till reach the confluence. The first panel shows green immunofluorescence for vimentin which is evident in the cell body and cytoplasmic processes in the cultured stellate cells. Red immunofluorescence for cygb is also positive. The second panel shows positive expression for α-smooth muscle actin. Both panels show blue immunofluorescence for DAPI, a cellular nuclei marker. Negative controls are showed on the bottom of each panel with all the markers used for the SC.

**If you need help for an experiment, just contact us, our experts will be pleased to assist you**

## CERTIFICATION:

*The viability and performance of the primary human liver endothelial cells provided depend primarily on the use of appropriate media and reagents, as well as the use of sterile plastics. Likewise, proper handling protocols must be followed. Please note that if these parameters are not carefully considered, the cellular response obtained in the assays may be lower than expected.*

| Name | Tittle | Signature | Cytes Biotechnologies, S.L. | Date |
|---|---|---|---|---|
| Pilar Sainz de la Maza | Quality Manager |  |  | 31/03/23 |