BIOMEDICUM
STEM CELL CENTER

IPS cell certification

Date: Sep 19, 2024

Cell Line: HEL348 clones HEL348.1 and HEL348.2

iPSC induction: Date of induction 06.11.2023. Fibroblasts were reprogramed using the CRISPRa method (PMID: 35063129)
Cell line origin: 69 KH+7
Investigator: *Tapanainen Juha/Khatun Masuma*

Culture Description: The iPSC lines were reprogrammed and cultured under feeder-free conditions. They were passaged using 0.5 mM EDTA on Matrigel-coated plates and maintained in E8 medium for over 10 passages.

Characterization:
i) iPSC morphology assessment: Monitoring typical iPSC colony morphology.
ii) Culture maintenance: Assessing the stability and survival of iPSC clones over ten passages.
iii) RT-PCR: Used to confirm the removal of transgene vectors.
iv) Mycoplasma test: Used to confirm the absence of mycoplasma.

Results:
i) All iPSC lines maintained typical human pluripotent stem cell morphology during extended culture.
ii) All clones survived and retained their morphological characteristics over multiple passages.
iii) Both HEL348 clones (HEL348.1 and HEL348.2) tested negative for the episomal vector.
iv) Mycoplasma test was negative for all lines.

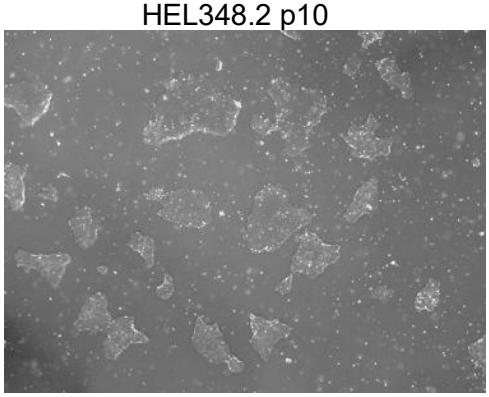
Interpretation:
Based on the analysis, both HEL348 clones (HEL348.1 and HEL348.2) meet the common criteria for iPSCs.

Recommendations:
- Use a Rho-kinase inhibitor (Y-27632) 24 hours after thawing to enhance cell survival.
- Fingerprinting has not been performed; it is important to match the produced iPSC lines to their corresponding donor cell lines.
- It is advisable to analyze the karyotype of the iPSC lines before using them in any experiments.
- Ensure compliance with the ISSCR Guidelines for Stem Cell Research and Clinical Translation (ISSCR Guidelines) for required characterizations, including morphology assessment, pluripotency verification, and genetic stability.

Contacts: *Diego Balboa (diego.balboa@helsinki.fi)*
Ras Trokovic (ras.trokovic@helsinki.fi)

A) Morphology assessment

HEL348.2 p10



B) OriP RT-PCR