Cell Type: Human Embryonic Stem Cells (H1)

Performance of Eppendorf Cell Culture Consumables (TC treated)

Background

The cultivation of human Pluripotent Stem Cells (hPSCs) is highly challenging as the maintenance of undifferentiated hPSCs requires certain culture conditions including an appropriate substrate to promote attachment and growth. The human Embryonic Stem Cell (hESC) line H1 - one of the most frequently used hESC lines reported in the literature by the end of 2008 [1] - was used to prove the suitability of Eppendorf Cell Culture Consumables for stem cell culture. The experiments were carried out in the Medical Research Council Laboratory of Molecular Biology in Cambridge, United Kingdom.

Table: Overview of culture media, coating and growth type of hESCs during testing

Image	Culture media	Coating	Growth type
A	Cellartis® DEF-CS™	Cellartis® DEF-CS COAT-1	Monolayer
B	StemCell™ Technologies mTeSR™1	Corning® Matrigel® Growth Factor Reduced, 85 µg per well	Colonies
C	Essential8 (internal/home made media composition according to Chen et al., 2011 [2])	Vitronectin, truncated recombinant human (VTN-N), 5 µg per well	Colonies

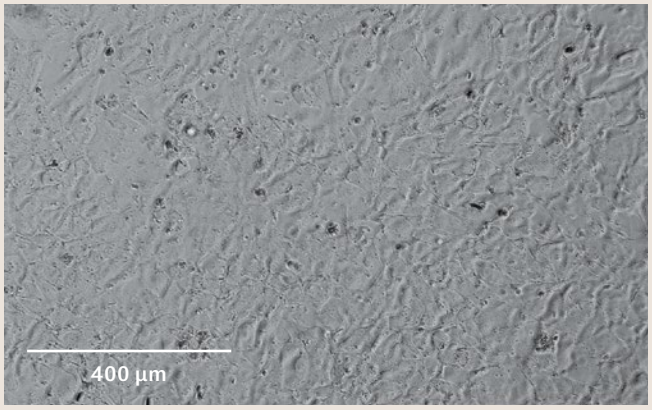
Procedure

Human Embryonic Stem Cells H1 were routinely cultured at 37°C with a humidified 5% CO_2 atmosphere and split twice a week at 80-85% confluency at a ratio of 1:6. All coatings were prepared according to manufacturer's instructions on TC treated Eppendorf Cell Culture Consumables.
For testing, hESCs were seeded in one of the appropriate culture media on pre-coated TC treated 6 well plates (Table). Cell confluence and morphology were monitored on day 3 post-seeding.

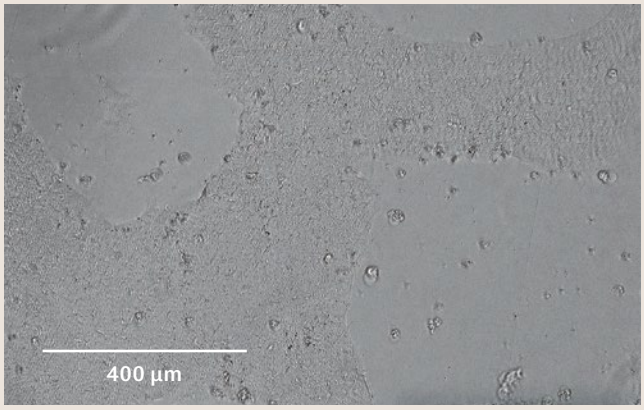
Results

hESCs observed on day 3 post-seeding displayed morphologies typical of feeder-free hESC cultures showing small and tightly packed cells grown in (A) a monolayer or (B-C) compact colonies with clean and defined edges without any sign of differentiation.

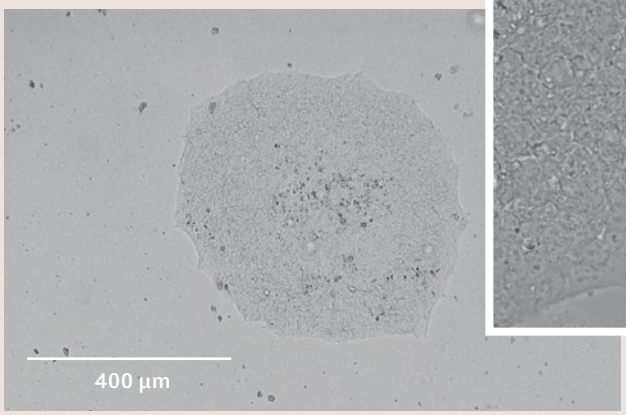
(A) DEF-CS COAT-1

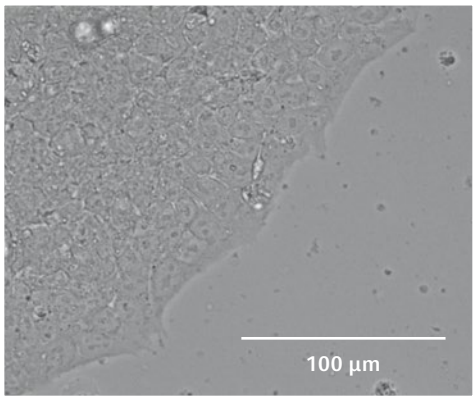


(B) Matrigel



(C) VTN-N





Representative light microscope images of hESCs on day 3 post-seeding on different coatings.

(A) Monolayer culture of hESCs on DEF-CS COAT-1
(B) hESC colonies on Matrigel, 85 µg per well
(C) hESC colonies on VTN-N, 5 µg per well

Conclusion

The TC treated surface of Eppendorf Cell Culture Consumables is compatible with several feeder-free stem cell culture systems and supports the correct colony morphology of human Embryonic Stem Cells. Eppendorf Cell Culture Consumables are for research use only.

Acknowledgement

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References

[1] Löser et al. (2010) Human Embryonic Stem Cell Lines and Their Use in International Research. Stem Cells, 28(2): 240–246.
[2] Chen et al. (2011) Chemically defined conditions for human iPS cell derivation and culture. Nat Methods, 8(5): 424-429.