FUJIFILM
Value from Innovation

Wako

For the further development of stem cell research

CultureSure™ CEPT Cocktail(1,000×)

CEPT is a cocktail of small molecules developed at the National Institutes of Health (NIH). It contains four components, and CEPT is an acronym for them: Chroman 1, Emricasan, Polyamines, and Trans-ISRIB.
Compared to existing methods, CEPT also improves cell viability in stem cell research, including embryoid body and organoid formation, single cell cloning, and genome editing using hPSCs.[1-5)]
*This product is manufactured and sold under license from NIH.

[References]

1) Chen, Y. *et al.* : *Nat. Methods*, **18 (5)**, 528 (2021).
2) Tristan, C.A. *et al.* : *Stem Cell Reports*, **16**, 3076 (2021).
3) Tristan, C.A. *et al.* : *Nat. Protoc.*, **18**, 58 (2022).
4) Deng, T. *et al.* : *Stem Cell Reports*, **18**, 1030 (2023).
5) Takeshi, W. *et al.* : *Regenerative Medicine*, **18(3)**, 219 (2023).

Features

- **Protects human ES/iPS cells from stresses including DNA damage, helping to maintain cell structure and function**
- **Filter-sterilized, ready-to-use cocktail solution**
- **One of the CultureSure series products that are tested for endotoxin contamination and are mycoplasma negative**

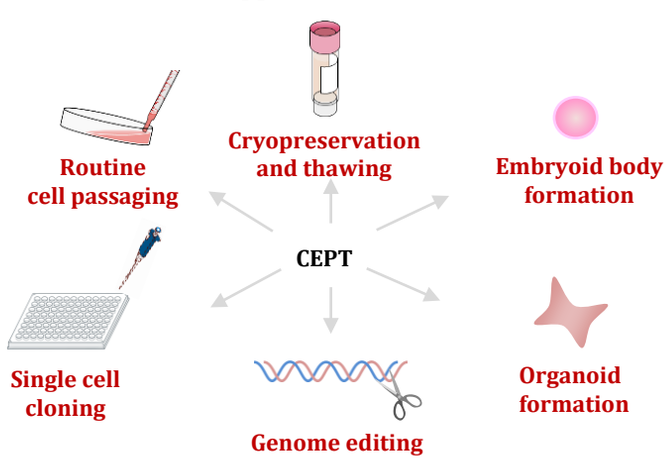
Analytical Data

- Concentration (HPLC): Passed
- Appearance: Liquid
- Endotoxin: Less than 3 EU/mL
- Tested for sterility
- Tested for negative mycoplasma contamination

Samples and Applications

Samples:
Human ES/iPS cells

Applications

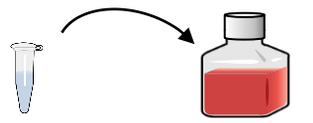


Wako Cat No.	Product Name	Grade	Package Size
033-26071	CultureSure™ CEPT Cocktail(1,000×)	for Cell Culture	300 µL

How to Use

Add 1/1,000 volume of this product to the culture medium and mix thoroughly before use.

Note: To avoid repeated freezing and thawing, it is recommended to aliquot in small volumes and freeze.

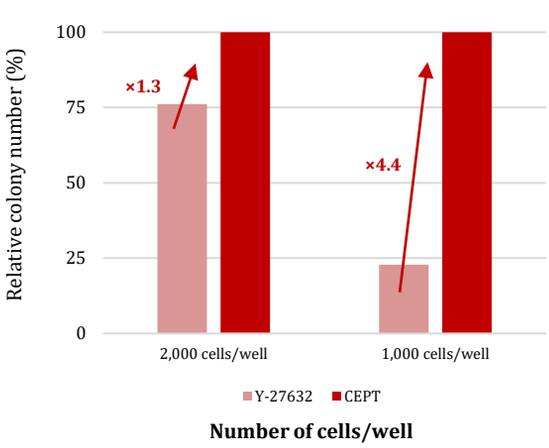


10 µL of CEPT Cocktail(1,000×) → 10 mL of hPSCs medium

Example of use: passaging of human iPS cells (For one 10-cm dish)

- Remove the medium from the culture dish.
- ← Add 5 to 10 mL of D-PBS (-) to the dish and rinse.
- Remove D-PBS (-).
- ← Add 2 to 5 mL of a cell dispersing agent.
- Allow to stand in an incubator set to 5% CO_2, 37 °C.
- ← Add 10 mL of hPSC medium+CEPT to the dish.
- Disperse the human iPS cell colonies into single cells by pipetting.
- ← Transfer the medium with dispersed cells into a tube.
- Centrifuge the tube for 5 min at 200 x g, room temperature, and remove the supernatant.
- ← Add 10 mL of hPSC medium+CEPT to the tube to suspend the cell pellet.
- Count cells.
- ← Seed an appropriate amount of human iPS cells in a new culture dish previously containing hPSC medium+CEPT.
- Culture in an incubator set to 5% CO_2, 37 °C.
- ← The following day, replace the medium with hPSC medium without CEPT.

Colony Formation Test when Cell Passaging

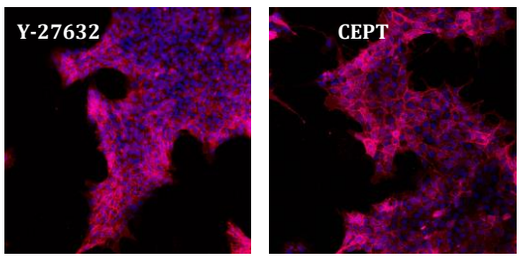


Cell strain	Human iPS cell 201B7 strain
Medium	StemSure® hPSC Medium Δ containing 35 ng/mL bFGF
Coating	Matrigel® hESC-Qualified Matrix
Number of seeded cells	2,000 cells/well, 1,000 cells/well (6 well plate)
Duration of Culture	8 days
Additives	Y-27632 or CEPT was added at the time of seeding. Cells were cultured without the addition from the following day onward.

Result

When the number of cells seeded was small (1,000 cells/well), more colonies were formed in the CEPT-supplemented cultures than in the Y-27632-supplemented cultures.

Undifferentiated State Maintenance



RED ：rBC2LCN-635 (human iPSC cell membrane staining)
BLUE ： DAPI (nuclear staining)

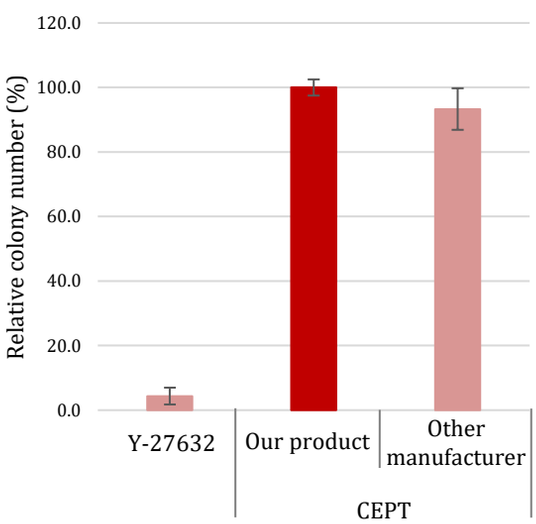
Note: BC2LCN is a recombinant lectin with high affinity for cell surface glycans of human ES/iPS cells.

Cell strain	Human iPS cell 201B7 strain
Medium	StemSure® hPSC Medium Δ containing 35 ng/mL bFGF
Coating	Matrigel® hESC-Qualified Matrix
Number of seeded cells	5,000 cells/well (12 well plate)
Duration of Culture	7 days
Additives	Y-27632 or CEPT was added at the time of seeding. Cells were cultured without the addition from the following day onward.

Result

No difference in cell morphology was observed between the addition of CEPT and Y-27632. The undifferentiated state was also maintained.

Colony Formation Test when Cell Passaging

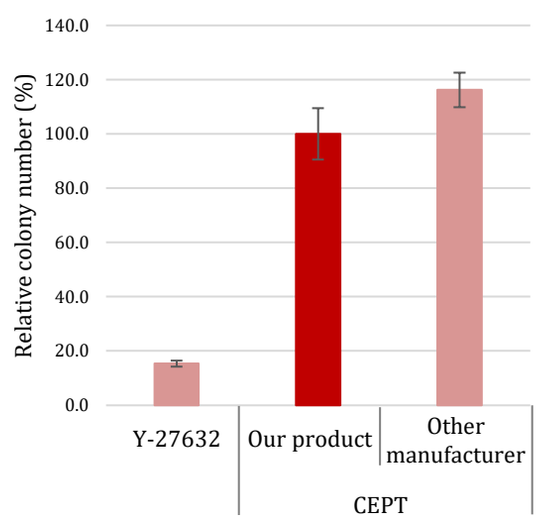


Cell strain	Human iPS cell 253G4 strain
Medium	StemSure® hPSC Medium Δ containing 40 ng/mL bFGF
Coating	Matrigel® hESC-Qualified Matrix
Number of seeded cells	1,000 cells/well（6 well plate）
Duration of Culture	7 days
Additives	Y-27632 or CEPT was added at the time of seeding. Cells were cultured without the addition from the following day onward.

Result

Human iPS cells were seeded in Y-27632 or CEPT-supplemented cultures when cell passaging. After 7 days of incubation, more colonies were formed in the CEPT-supplemented cultures than Y-27632. In addition, our product showed the same performance as other manufacturer.

Colony Formation Test when Thawing Frozen Cells



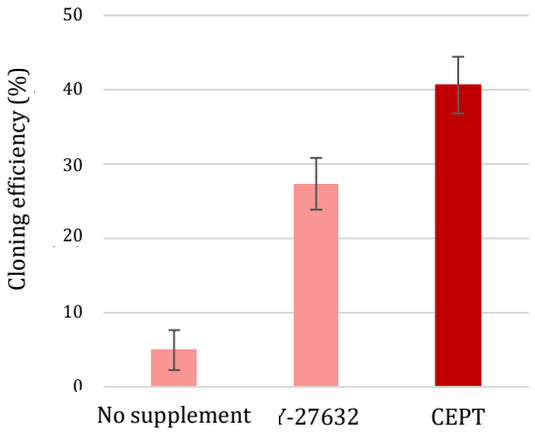
Cell strain	Human iPS cell 253G4 strain
Medium	StemSure® hPSC Medium Δ containing 40 ng/mL bFGF
Coating	Matrigel® hESC-Qualified Matrix
Number of seeded cells	1,000 cells/well（6 well plate）
Duration of Culture	7 days
Additives	Y-27632 or CEPT was added at the time of seeding. Cells were cultured without the addition from the following day onward.

Result

Frozen human iPS cells were thawed and seeded in Y-27632 or CEPT-supplemented cultures. After 7 days of incubation, more colonies were formed in the CEPT-supplemented cultures than Y-27632. In addition, our product showed the same performance as other manufacturer.

Efficacy for Single Cell Cloning

Human iPS cells were seeded in each well of the 96-well plate so that they would be single-cells, and after 9 days of incubation, the number of wells that had formed colonies was counted.



Cell strain	Human iPS cell 201B7 strain
Medium	StemSure® hPSC Medium Δ containing 35 ng/mL bFGF
Coating	Matrigel® hESC-Qualified Matrix
Number of seeded cells	1 cell/well（96well plate）
Duration of Culture	9 days
Additives	Y-27632 or CEPT was added at the time of seeding. Cells were cultured without the addition from the following day onward.

Result

CEPT cocktail was shown a remarkable cytoprotective effect under high-stress conditions and significantly increase the cloning efficiency in single-cell cloning.

Related Products

Wako Cat No.	Product Name	Grade	Package Size
039-24591	CultureSure® 10mmol/L Y-27632 Solution, Animal-derived-free	for Cell Culture	300 uL
035-24593			1 mL
038-24681	CultureSure® 10mmol/L CHIR99021 DMSO Solution, Animal-derived-free	for Cell Culture	300 uL
033-24631	CultureSure® 5mmol/L SB431542 DMSO Solution, Animal-derived-free	for Cell Culture	1 mL

Please check our website for more details.

wako cept Search

https://labchem-wako.fujifilm.com/us/category/03127.html

Please check our catalog for ES/iPS cells and regenerative medicine research.

https://labchem-wako-pages.fujifilm.com/US-SmallMolecules-Catalog-Download.html

FUJIFILM Wako Laboratory Chemicals site
https://labchem-wako.fujifilm.com

FUJIFILM Wako Pure Chemical Corporation
1-2, Doshomachi 3-Chome, Chuo-ku, Osaka 540-8605, Japan
ffwk-cservise@fujifilm.com

FUJIFILM Irvine Scientific
1830 E. Warner, Avenue, Santa Ana, CA 92705-5505, U.S.A.
Toll-Free (U.S. only): +1 800 577 6097
Tel: +1 949 261 7800 Fax: +1 949 261 6522
fisilssupport@fujifilm.com

FUJIFILM Wako Chemicals Europe GmbH
Fuggerstr 12, 41468 Neuss, Germany
Tel: +49 2131 311 0 Fax: +49 2131 311 100
labchem_wkeu@fujifilm.com

FUJIFILM Wako (Guangzhou) Trading Corporation
Room 3003, 30/F., Dong Shan Plaza 69, Xian Lie Zhong Road, Guangzhou, 510095, China
Tel: +86-20-8732-6381(Guangzhou) Tel: +86-21-6288-4751(Shanghai)
Tel: +86-10-6413-6388(Beijing)
wkgz.info@fujifilm.com

FUJIFILM Wako Chemicals (Hong Kong) Limited
Room 1111, 11/F, International Trade Centre, 11-19 Sha Tsui Road, Tsuen Wan, N.T., Hong Kong
Tel: +852-2799-9019 Fax: +852-2799-9808
wkhk.info@fujifilm.com

25200学31