HE100 medium

Chemically defined medium

Description

HE100 medium has been developed for the growth of human embryonic kidney (HEK) 293 cells, such as Expi293F, 293-F, 293T, or VPC cells, and the high production of recombinant proteins or the transfection assay in serum-free culture. HE100 medium is a chemically defined, serum-free, protein-free, animal origin-free medium that contains no protein, hydrolysates, or components of unknown composition.

(Storage; 2℃ to 8℃ / Protect from light)

Culture conditions

Cell line: 293 cells
Culture type: Adhesive
Culture vessels: Flask, plate, dish, or culture bag, etc.
Incubate atmosphere: Humidified atmosphere of 5–8% CO_2 in air
Temperature range: 36℃ to 38℃

Prepare medium

HE100 medium requires supplementation with L-glutamine or L-alanyl-L-glutamine.

1 Add 200 mM L-glutamine or L-alanyl-L-glutamine, 2–8 mM final concentration, to the medium.
2 HE100 medium contains no antibiotics. Please supply to the medium as necessary.

Thaw and Subculture method

For Static Culture (T75 Flasks)

1 Add 200 mM L-glutamine (4 mM final concentration, 20 mL/L) to the medium before use.
2 Thaw 293 cells in a water bath and transfer into a 15-mL tube containing 10 mL of HE100 medium.
3 Resuspend with 10 mL of HE100 medium, count cells and determine cell viability.
4 Transfer cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) into a T75 flask containing 25 mL of HE100 medium and incubate at 37℃.
5 On the second day culture, harvest cells by pipetting with a 50-mL tube. **Trypsin may be used to harvest cells. Remove the trypsin with trypsin inhibitor to prevent cell damage and death.**
6 Resuspend with 10 mL of HE100 medium and determine the viable cell density.
7 Transfer cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) into a T75 flask containing 25 mL of HE100 medium and incubate at 37℃.
8 On the fourth day culture, harvest cells with a 50-mL tube, and determine the viable cell density.
9 Subculture cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) every 4 days (3–5 days)

with fresh HE100 medium.

10 For your experiments before using, subculture 293 cells a minimum of three times to allow them to recover from cryopreservation.

For Static Culture (100-mm Dishes)

1 Add 200 mM L-glutamine (4 mM final concentration, 20 mL/L) to the medium before use.
2 Thaw 293 cells in a water bath and transfer into a 15-mL tube containing 10 mL of HE100 medium.
3 Resuspend with 10 mL of HE100 medium, count cells and determine cell viability.
4 Transfer cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) into a 100-mm dish containing 20 mL of HE100 medium and incubate at 37℃.
5 On the second day culture, harvest cells by pipetting with a 50-mL tube. **Trypsin may be used to harvest cells. Remove the trypsin with trypsin inhibitor to prevent cell damage and death.**
6 Resuspend with 10 mL of HE100 medium and determine the viable cell density.
7 Transfer cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) into a 100-mm dish containing 20 mL of HE100 medium and incubate at 37℃.
8 On the fourth day culture, harvest cells with a 50-mL tube, and determine the viable cell density.
9 Subculture cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) every 4 days (3–5 days) with fresh HE100 medium.
10 For your experiments before using, subculture 293 cells a minimum of three times to allow them to recover from cryopreservation.

Adaptation method to HE100 medium

We recommend both direct and sequential adaptation method, adapting 293 cells to HE100 medium. It is critical that the growth rate is in mid-logarithmic phase before adaptation culture.

For direct adaptation (T75 Flasks)

1 Add 200 mM L-glutamine (4 mM final concentration, 20 mL/L) to the medium before use.
2 Harvest cells and ensure that the growth rate is in mid-logarithmic phase.
3 Transfer cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) into a T75 flask containing 25 mL of HE100 medium and incubate at 37℃.
4 On the fourth day culture, harvest cells by pipetting with a 50-mL tube and determine the viable cell density. **Trypsin may be used to harvest cells. Remove the trypsin with trypsin inhibitor to prevent cell damage and death.**
5 Subculture cells at a seeding density of 3 x 10^5 cells/mL (2–4 x 10^5 cells/mL) every 4 days (3–5 days) with fresh HE100 medium.
6 Continue to subculture cells as necessary every 4 days (3–5 days) with fresh HE100 medium until consistent growth is achieved.

Cryopreservation

1 Prepare the cryopreservation medium of 90% HE100 medium and 10% DMSO.
2 Harvest cells and resuspend at a cell density of 5–10 x 10^6 cells/mL with the fresh cryopreservation medium.
3 Transfer 293 cells into cryovials.
4 Achieve cryopreservation following standard procedures, do not directly put into liquid nitrogen.
5 Transfer frozen cells to liquid nitrogen.

Other information

For Research Use Only. Not for use in diagnostic procedures. This product is sold for research and development purposes only. It is not for any human or animal therapeutic or clinical diagnostic use. It is not intended for food, drug, household, agricultural, or cosmetic use. Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

Related product

＜ Transfection System ＞

Gxpress 293 Transfection & Medium Kit	GX293-MAK-0010
Gxpress 293 Transfection & Medium Kit II	GX293-MK-0010
Gxpress 293 Transfection Kit	GX293-RK-0010
Gxpress 293 TF Reagent	GX293-TF-0010
Gxpress 293 Enhancer	GX293-EN-0010

＜ Chemically Defined Medium ＞

HE100 medium	HE100-0010	Adhesive culture
HE150 medium	HE150-0005	Cloning assay
HE200 medium	HE200-0010	Suspension culture
HE300 medium	HE300-0010	Suspension culture
HE300AZ medium*	HE300AZ-0010	Suspension culture
HE400 medium	HE400-0010	Suspension culture
HE400AZ medium*	HE400AZ-0010	Suspension culture
Gxpress 293 Feed medium	GX293-FD-0010	Fed-Batch culture

* Ready-to-use medium with L-alanyl-L-glutamine