

AdvanceSTEM Low Osmo DMEM

HYCLONE MEDIA AND SUPPLEMENTS

HyClone™ AdvanceSTEM Low Osmo DMEM is developed to support the growth and maintenance of murine embryonic stem cells (Fig 1). The product's osmolality has been optimized to approximate murine embryonic tissue.

Features of AdvanceSTEM Low Osmo DMEM include:

- Developed to support growth and maintenance of murine embryonic stem cells
- Formulated to closely approximate the osmolality of embryonic tissues
- Specifically formulated to maintain physiological pH at 37°C and 5% CO₂

Specifications

AdvanceSTEM Low Osmo DMEM contains HEPES, β-mercaptoethanol, and non-essential amino acids (NEAA). The formulation does not contain L-glutamine to allow users to supplement at the L-glutamine concentration of their choice. This basal medium requires supplementation with fetal bovine serum (FBS) or serum replacement.

Suggested preparation

Recommended supplementation is 15% FBS or 20% HyClone AdvanceSTEM Serum Replacement. AdvanceSTEM Low Osmo DMEM does not contain leukemia inhibitory factor (LIF). For the culture of mouse embryonic stem cells (mESC), supplementation with LIF is required with this formulation when FBS or serum replacements are used (Fig 2).



Fig 1. AdvanceSTEM Low Osmo DMEM.

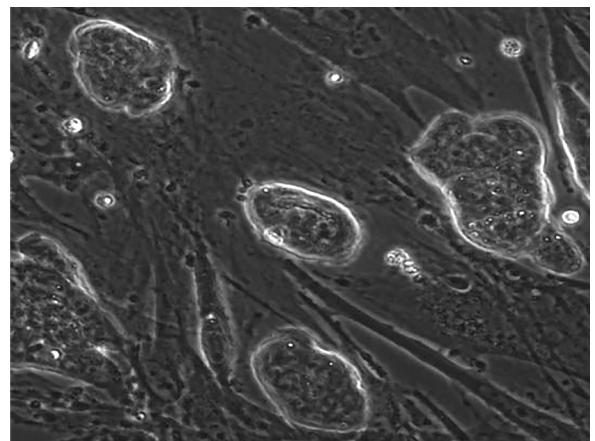


Fig 2. C57BL/6J mESCs growing on mouse embryonic fibroblast (MEF) feeder cells. The mESCs were derived and propagated in AdvanceSTEM Low Osmo DMEM and AdvanceSTEM Serum Replacement. Photo courtesy of Primogenix, Inc.

General culture recommendations

1. Cultures should be incubated at 37°C in a 5% CO₂ environment.
2. To maintain mESCs in an undifferentiated state, examine cells daily under a phase contrast microscope and passage if necessary. Passage cells before signs of differentiation appear.
3. Change medium daily, use medium supplemented as follows:
 - a. 20% AdvanceSTEM Serum Replacement supplementation (Table 1), or 15% FBS supplementation (Table 2).
 - b. LIF supplementation (1000 U/mL) is required.
 - c. L-glutamine supplementation is required.
4. Trypsinize and passage cells every other day at approximately a ratio of 1 to 5 in a new flask of MEF cells. Roughly 1/5 of the trypsinized cells are passaged to a new flask of inactivated MEF cells, the remaining cells are either frozen or used experimentally as needed.
 - a. When using FBS, follow trypsinization methods outlined in the procedure Mouse embryonic feeder cell protocol: subculturing MEF cells (29154593AA).
 - b. When using serum replacement, follow trypsinization methods outlined in the data file AdvanceSTEM Serum Replacement, Section General culture recommendations (29153879 AA).

Note: subculturing of mESCs by trypsinization is a proven method for dissociation of mESCs colonies. However, care should be taken when passaging in medium containing serum replacements, as serum replacements do not contain the trypsin inhibitors found in FBS, making it necessary to remove the trypsin before plating the cells.

Table 1. Preparation of 200 mL AdvanceSTEM Low Osmo DMEM supplemented with 20% AdvanceSTEM Serum Replacement

Product	Amount for 200 mL	Product code
HyClone AdvanceSTEM Low Osmo DMEM	158 mL	SH30870.01, SH30870.02
HyClone AdvanceSTEM Serum Replacement	40 mL	SH30874.01, SH30874.02
HyClone AdvanceSTEM ES Qualified L-glutamine 200 mM	2.0 mL	SH30852.01
ESGRO™-Mouse LIF (Millipore)	20 µL	ESG1107

Store at 2°C to 8°C and discard any unused medium after 2 weeks.

Table 2. Preparation of 200 mL AdvanceSTEM Low Osmo DMEM supplemented with 15% FBS

Product	Amount for 200 mL	Product code
HyClone AdvanceSTEM Low Osmo DMEM	168 mL	SH30870.01, SH30870.02
HyClone FBS-ES Qualified	30 mL	SH30070.03E
HyClone AdvanceSTEM ES Qualified L-glutamine 200 mM	2.0 mL	SH30852.01
ESGRO-Mouse LIF (Millipore)	20 µL	ESG1107

Store at 2°C to 8°C and discard any unused medium after 2 weeks.

Quality control testing

Quality control test specifications are listed in Table 3.

Table 3. Test specifications*

Test	Specification
Appearance	Clear reddish solution
Osmolality	270–290 mOsm/kg
pH	6.9–7.3
Sterility	No growth (bacteria or fungi)
Endotoxin	≤ 1 EU/mL
Application	Plating efficiency ≥ 15%

* Refer to certificate of analysis for actual results.

Related products

Classic culture conditions for mESCs include co-culturing with primary MEF cells in medium containing FBS and supplements. The following products are used in the culture of mESCs.

HyClone ES Screened FBS

For research involving embryonic stem cells (ESCs), it is critical to maintain the cells in their undifferentiated state. ES Screened FBS has been screened for the ability to promote the rapid growth of ES cells while retaining their pluripotent state, eliminating the need for customers to prescreen serum lots.

ES Qualified Stem Cell Products

HyClone AdvanceSTEM line of stem cell culture products has been developed specifically for applications in stem cell research. The line includes numerous products designed to support a wide range of applications, from the growth and maintenance of murine embryonic and human adult somatic stem cells to the directed differentiation of mesenchymal stem cells into adipocytes, chondrocytes, and osteocytes. In addition to AdvanceSTEM Low Osmo DMEM, the range of products include:

- AdvanceSTEM Serum Replacement
- AdvanceSTEM ES Qualified L-glutamine
- AdvanceSTEM ES Qualified Non-Essential Amino Acids
- AdvanceSTEM ES Qualified HEPES
- AdvanceSTEM ES Qualified PBS
- AdvanceSTEM ES Qualified Dulbecco's Phosphate Buffered Saline (DPBS)
- AdvanceSTEM IMDM4SC without L-glutamine
- AdvanceSTEM DMEM4SC with 4.5 mg/L glucose, without L-glutamine and sodium pyruvate

Ordering information

Product	Size	Product code
HyClone AdvanceSTEM Low Osmo DMEM Without L-glutamine	500 mL	SH30870.01
	1000 mL	SH30870.02

Related products	Size	Product code
HyClone AdvanceSTEM Serum Replacement, Frozen	50 mL	SH30874.01
	100 mL	SH30874.02
	500 mL	SH30874.03
HyClone ES Screened FBS	500 mL	SH30070.03E
HyClone AdvanceSTEM ES Qualified L-glutamine	100 mL	SH30852.01
HyClone AdvanceSTEM ES Qualified Non-Essential Amino Acids	100 mL	SH30853.01
HyClone AdvanceSTEM ES Qualified HEPES	100 mL	SH30851.01
HyClone AdvanceSTEM ES Qualified PBS	1000 mL	SH30850.03
HyClone AdvanceSTEM ES Qualified Dulbecco's Phosphate Buffered Saline (DPBS)	500 mL	SH30850.02
	1000 mL	SH30850.03
HyClone AdvanceSTEM IMDM4SC Without L-glutamine	500 mL	SH30822.01
	1000 mL	SH30822.02
HyClone AdvanceSTEM DMEM4SC With 4.5 mg/L glucose Without L-glutamine and sodium pyruvate	500 mL	SH30824.01
	1000 mL	SH30824.04

* Certificates of analysis, product inserts, and protocols can be found at www.cytiva.com/hyclonecerts

cytiva.com/hyclone

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