HL-1™ Supplement

HL-1™Supplement is a chemically-defined medium additive that can be used to replace serum or significantly reduce its concentration in a variety of basal media. It contains less than 30µg protein/ml when diluted 1:100 in medium and does not contain bovine serum albumin or other undefined protein ingredients. HL-1™ Supplement, when added to most basal media, will support the growth of mouse hybridoma cells and certain other differentiated cells of lymphoid

Instructions for Use

1. Aseptically add 10 ml of HL-1™ Supplement to 1 litre of sterile basal medium of choice. The recommended basal media are:
a.1:1 mixture of Dulbecco's Modified Eagle Medium (DMEM) and Ham's F12 (high glucose)
b. RPMI-1640
c. Iscove's Modified Dulbecco's Medium (IMDM)
d. DMEM containing up to 1% foetal bovine serum (FBS)
e. Other enriched media containing amino acids.

NOTE: The medium should also be supplemented with L-glutamine (2-4mM), sodium Pyruvate (1-2mM), and sodium bicarbonate (2.2 g/L or 1.2 g/L), if not already present. The pH of the basal medium should be the specification recommended by the manufacturer (typically pH 7.2), as the pH will not change after the addition of the HL-1™ Supplement. Since HL-1™ Supplement is sterile, filtration of sterile medium following its addition is not necessary.

Antibiotics and pH stabilizing buffers such as HEPES or MOPS may be added at the user's discretion. It should be noted, however, that these additives may affect cell growth since some cells appear to become more sensitive to these agents under serum-free conditions.

Adaptation of Serum-dependent Cells

Weaning cell lines from serum-containing media is required if serum-free culture is desired. The following protocol will aid adaptation to a serum-free environment using HL-1™Medium and gradual reduction of serum concentration (see note below)

Week 1: Reduce the serum concentration to 5% FBS in HL-1™Medium.

Week 2: Reduce the serum concentration to 2% FBS in HL-1™Medium.

Week 3: Reduce the serum concentration to 1% FBS in HL-1™Medium.

Week 4: Reduce the serum concentration to 0.5% FBS in HL-1™Medium.

Week 5: Reduce the serum concentration to 0.25% FBS in HL-1™ Medium.

Week 6: Eliminate FBS in HL-1™Medium and culture cells in serum-free HL-1™Medium.

NOTE: At each reduction stage, cells may show evidence of an initial lag in growth rate. Pass the cells three times per week during the adaptation period, seeding at a density of 1-2 x 10^5cells/ml. Do not allow

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densities to exceed 8-10 x 10^5cells/ml. Upon reduction to the 0.5% serum concentration, a greater lag in the growth rate may be observed. Under these conditions, a higher seeding density and less frequent passaging may be required until cells resume their normal growth characteristics.

HL-1™ Supplement Cell Testing

HL-1™ has been successfully tested on the following cells types:

Transformed & Established Cell Lines

BB88	murine	erythroid (leukaemia)
U937	human	macrophage
P815	murine	macrophage
P388D1	murine	macrophage
WeHi3	murine	monocyte
JLS-V5	murine	spleen cell
RaJi	human	B lymphoblastic
GCL2	hamster/ mouse	B lymphoma X Normal B
70Z-3	murine	Pre-B lymphoma
70Z/3.12	murine	B lymphoma
S49 and variants	murine	T lymphoma
RAW309F1.1	murine	T lymphoma
WeHi7	murine	T lymphoma
L5178Y	murine	(DBA/2)lymphoma
I-10	murine	Leydig -tumour
MCF-7 (NIH)	human	breast carcinoma
MCF-7 (MCF)	human	breast carcinoma
NIH ZR-75	human	breast carcinoma
COLO 302 HSR	human	colon carcinoma
J8	human	bladder carcinoma
SW 1738	human	bladder carcinoma

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SW780	human	bladder carcinoma
EL4	murine	T lymphoma
RL1	murine	T lymphoma
BW5147.3	murine	T lymphoma
LBRM-33	murine	T lymphoma
Friend leukaemia	murine	lymphoid
CCL 213	human	Burkitt lymphoma
C91/PL	human	T lymphoma
Undesignated	human	astrocytoma
Undesignated	human	hepatoma

Transformed & Established Cell Lines

VERO	African green monkey	fibroblast
MDCK	canine	Madin Darby canine kidney
MOLT-3	human	acute lymphoblastic leukaemia
MOLT-4	human	acute lymphoblastic leukaemia
NAMALWA	human	Burkitt lymphoma
C57BL6	murine	(C57 X DBA) embryo
CHO K1	hamster	Chinese hamster ovary (epith.-like)
THP-1	human	monocytic leukaemia

Hybridomas

HB44	murine	Sp2/0-Ag14
HB45	murine	Sp2/0-Ag14
HB56	murine	NS-1
HB59	murine	NS-1
HB60	murine	P3X63Ag 8.653
53-7.313	murine	NS-1
MI/9.3.4HL-2	murine	NS-1

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8A1	human	CLLC
MI/70.15.1	murine	NS-1
ARB	murine	hybridoma
P3U	murine	P3X63Ag 8.653
TIB 175	rat/mouse	S194
TIB 104	rat/mouse	NS-1
TIB 105	rat/mouse	NS-1
TIB 109	rat/mouse	NS-1
TIB 128	rat/mouse	NS-1
TIB 166	rat/mouse	NS-1
TIB 168	rat/mouse	NS-1
RS	rat/mouse	P3X63Ag 8.653
BCS12	murine	P3X63Ag 8.653
BCS 2002	murine	P3X63Ag 8.653
Undesignated	human	WI-L2-729-HF2
Undesignated	human	LICR-LON-HMY2
Undesignated	murine	NS-1
Undesignated	murine	P3X63Ag 8.653

Primary Cells

Human peripheral blood mononuclear

Mink lymphocytes

Human foetal adrenal

Human blood monocytes

Human peripheral blood T lymphocytes

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Presentation

Product code	Description	Size
LSG-1061A x 2	HL-™Medium; chemically defined, serum-free medium	2 x 500mL
LSG-1062E	HL-1™Supplement; chemically defined, serum-free supplement (100X)	10mL

Storage and Stability

HL-1™Supplement should be stored at 15-30°C. The shelf life of HL-1™Supplement is 1 year from the date of manufacture. Once diluted into basal medium it should be used within 45 days.

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THESE PRODUCTS ARE FOR RESEARCH USE ONLY.
Not approved for human or veterinary use, for application to humans or animals, or for use in clinical or *in vitro* diagnostic procedures.

Support

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