

## MCDB Media

MCDB media were designed for the low-protein or serum-free growth of specific cell types using hormones, growth factors, trace elements or low levels of dialyzed fetal bovine serum protein (FBSP). Each MCDB medium was formulated (qualitatively and quantitatively) to provide a defined and optimally balanced nutritional environment that selectively promoted growth of a specific cell type. MCDB 105 and 110 are modifications of MCDB 104 medium, optimized for long-term survival and rapid clonal growth of human diploid fibroblast-like cells (WI-38, MRC-5, IMR-90) and of low-passage human foreskin fibroblasts using FBSP or hormone and growth factor supplements. MCDB 151, 201 and 302 are modifications of Ham's nutrient mixture F-12, designed for the growth of human keratinocytes, clonal growth of chicken embryo fibroblasts and chinese hamster ovary (CHO) cells using low levels of FBSP, extensive trace elements or no serum protein.

	<b>M 6395</b>	<b>M 6520</b>	<b>M 6645</b>	<b>M 8537</b>
	(105)	(110)	(151)	(131)
<b>COMPONENT</b>	g/L	g/L	g/L	g/L
<b>INORGANIC SALTS</b>				
NH <sub>4</sub> VO <sub>3</sub>	0.00000585	0.00000585	—	0.000006
CaCl <sub>2</sub> •2H <sub>2</sub> O	0.147	0.147	0.004411	0.2352
CuSO <sub>4</sub> •5H <sub>2</sub> O	0.0000025	0.0000025	0.0000025	0.0000012
FeSO <sub>4</sub> •7H <sub>2</sub> O	0.00139	0.00139	0.000417	0.000278
MgCl•6H <sub>2</sub> O	—	—	0.122	—
MgSO <sub>4</sub> (anhyd)	0.12038	0.12038	—	1.204
MnSO <sub>4</sub>	0.00000151	0.00000151	—	0.0000002
(NH <sub>4</sub> ) <sub>2</sub> MO <sub>4</sub> •4H <sub>2</sub> O	0.00000124	0.00000124	—	0.0000037
NiCl <sub>2</sub> •6H <sub>2</sub> O	0.00000012	0.00000012	—	0.0000001
KCl	—	0.37275	0.11183	0.2982
KH <sub>2</sub> PO <sub>4</sub> (anhyd)	0.40827	—	—	—
Na•Acetate (anhyd)	—	—	0.30153	—
NaCl	6.546	6.546	7.599	6.4284
NaSiO <sub>3</sub> •9H <sub>2</sub> O	0.0001421	0.000142	—	0.002842
Na <sub>2</sub> HPO <sub>4</sub> (anhyd)	—	0.42594	0.284088	0.071
Na <sub>2</sub> SeO <sub>3</sub>	0.000005187	0.0000038	—	0.0000052
SnCl <sub>2</sub> •2H <sub>2</sub> O	0.000000113	0.000000113	—	—
ZnSO <sub>4</sub> •7H <sub>2</sub> O	0.000144	0.000144	0.000863	0.0000003
<b>AMINO ACIDS</b>				
L-Alanine	0.00891	0.00891	0.00891	0.00267
L-Arginine•HCl	0.2107	0.2107	0.2107	0.06321
L-Asparagine•H <sub>2</sub> O	0.015	0.015	0.015	0.01501
L-Aspartic Acid	0.01331	0.01331	0.00399	0.01331
L-Cysteine•HCl•H <sub>2</sub> O	0.00878	0.00878	0.04204	0.03512
L-Glutamic Acid	0.01471	0.01471	0.01471	0.004413
L-Glutamine	0.3653	0.3653	0.8772	1.461
Glycine	0.00751	0.02252	0.00751	0.00225
L-Histidine•HCl•H <sub>2</sub> O	0.02097	0.02097	0.01677	0.04192
L-Isoleucine	0.00394	0.00394	0.001968	0.0656
L-Leucine	0.01312	0.01312	0.0656	0.1312
L-Lysine•HCl	0.03654	0.03654	0.01827	0.1826
L-Methionine	0.00448	0.00448	0.004476	0.01492
L-Phenylalanine	0.00496	0.00496	0.004956	0.03304
L-Proline	0.03453	0.03453	0.03453	0.01151
L-Serine	0.01051	0.01051	0.06306	0.03153
L-Threonine	0.01191	0.01191	0.01191	0.01191
L-Tryptophan	0.00204	0.00204	0.00306	0.00408
L-Tyrosine•2Na•2H <sub>2</sub> O	0.00784	0.00784	0.00392	0.02252
L-Valine	0.01172	0.01172	0.03513	0.1171

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MCDB Media continued

	<b>M 6395</b>	<b>M 6520</b>	<b>M 6645</b>	<b>M 8537</b>
	(105)	(110)	(151)	(131)
<b>COMPONENT</b>	g/L	g/L	g/L	g/L
<b>VITAMINS</b>				
D-Biotin	0.000007339	0.00000733	0.0000146	0.0000073
Choline Chloride	0.01396	0.01396	0.01396	0.01396
Folic Acid	—	—	0.00079	—
Folinic Acid•Ca	0.000000512	0.000000602	—	0.0005115
myo-Inositol	0.01802	0.01802	0.01802	0.007208
Niacinamide	0.00611	0.00611	0.0000366	0.006105
D-Pantothenic Acid•½Ca	0.000238	0.000238	0.000238	0.011915
Pyridoxine•HCl	0.0000617	0.0000617	0.0000617	0.002056
Riboflavin	0.000113	0.000113	0.0000376	0.0000038
Thiamine•HCl	0.000337	0.000337	0.000337	0.003373
Vitamin B-12	0.000136	0.000136	0.000407	0.0000136
<b>OTHER</b>				
Adenine•HCl	0.00172	0.00172	0.03088	0.0001716
D-Glucose	0.72064	0.72064	1.081	1.0
HEPES	5.958	5.958	6.6	—
Linoleic Acid	0.0000028	—	—	—
Phenol Red•Na	0.001242	0.001242	0.001242	0.0124212
Putrescine•2HCl	0.000000161	0.000000161	0.0001611	0.0000002
Pyruvic Acid•Na	0.11	0.11	0.055	0.11
Thioctic Acid	0.00000206	0.00000206	0.0002063	0.0000021
Thymidine	0.0000727	0.0000727	0.000727	0.0000242
<b>ADD</b>				
NaHCO <sub>3</sub>	N/A	N/A	1.176	1.18
Grams of powder required to prepare 1 L	14.9	15.3	17.7	11.7

**REFERENCES**

1. Peehl, DM. and Ham, R.G., (1980). In Vitro, 16:526.
2. Bettger, W.J., et al., (1981) Rapid Clonal Growth and Serial Passage of Human Diploid Fibroblasts in a Lipid-Enriched Synthetic Medium Supplemented with Epidermal Growth Factor, Insulin, and Dexamethasone. Proc. Natl. Acad. Sci., USA, 78:9, 5588-5592.

MCDB Media continued

	<b>M 7403</b>	<b>M 6770</b>	<b>M 2021</b>
	(153)	(201)	(302)
<b>COMPONENT</b>	g/L	g/L	g/L
<b>INORGANIC SALTS</b>			
NH <sub>4</sub> VO <sub>3</sub>	0.00000585	0.00000006	0.0000117
CaCl <sub>2</sub> •2H <sub>2</sub> O	0.004411	0.294	0.08821
CuSO <sub>4</sub> •5H <sub>2</sub> O	0.0000275	0.0000025	0.000025
FeSO <sub>4</sub> •7H <sub>2</sub> O	0.00139	0.001668	0.000834
MgCl•6H <sub>2</sub> O	0.122	—	0.122
MgSO <sub>4</sub> (anhyd)	—	0.18057	—
MnSO <sub>4</sub>	0.00000151	0.00000075	0.00000151
(NH <sub>4</sub> ) <sub>2</sub> MO <sub>4</sub> •4H <sub>2</sub> O	0.0000124	0.00000618	0.0000124
NiCl <sub>2</sub> •6H <sub>2</sub> O	0.0000012	0.000000012	—
KCl	0.11183	0.22365	0.22365
KH <sub>2</sub> PO <sub>4</sub> (anhyd)	—	—	—
Na•Acetate (anhyd)	0.30153	—	—
NaCl	7.599	7.597	7.599
NaSiO <sub>3</sub> •9H <sub>2</sub> O	0.000142	0.000142	—
Na <sub>2</sub> HPO <sub>4</sub> (anhyd)	0.284088	0.07099	0.14198
Na <sub>2</sub> SeO <sub>3</sub>	0.0000038	0.000000865	0.00000173
SnCl <sub>2</sub> •2H <sub>2</sub> O	0.00000113	—	—
ZnSO <sub>4</sub> •7H <sub>2</sub> O	0.000144	0.000028744	0.000863
<b>AMINO ACIDS</b>			
L-Alanine	0.00891	0.00891	0.00891
L-Arginine•HCl	0.2107	0.0632	0.2107
L-Asparagine•H <sub>2</sub> O	0.015	0.150	0.015
L-Aspartic Acid	0.00399	0.01331	0.01331
L-Cysteine•HCl•H <sub>2</sub> O	0.04204	0.03513	0.01756
L-Glutamic Acid	0.01471	0.01471	0.01471
L-Glutamine	0.8772	0.14615	0.4386
Glycine	0.00751	0.00751	0.00751
L-Histidine•HCl•H <sub>2</sub> O	0.01677	0.02097	0.02097
L-Isoleucine	0.001968	0.01312	0.00394
L-Leucine	0.0656	0.03935	0.01312
L-Lysine•HCl	0.01827	0.03654	0.03654
L-Methionine	0.00448	0.00448	0.00448
L-Phenylalanine	0.00496	0.00496	0.00496
L-Proline	0.03453	0.00576	0.03453
L-Serine	0.06306	0.03153	0.01051
L-Threonine	0.01191	0.03574	0.01191
L-Tryptophan	0.00306	0.00613	0.00204
L-Tyrosine•Na	0.00341	0.01135	0.007896
L-Valine	0.03513	0.03513	0.01172

Formulas continued on next page

MCDB Media continued

	<b>M 7403</b>	<b>M 6770</b>	<b>M 2021</b>
	(153)	(201)	(302)
<b>COMPONENT</b>	g/L	g/L	g/L
<b>VITAMINS</b>			
D-Biotin	0.0000146	0.00000733	0.00000733
Choline Chloride	0.01396	0.01396	0.01396
Folic Acid	0.00079	—	0.001324
Folinic Acid•Ca	—	0.00000512	—
myo-Inositol	0.01802	0.01802	0.01802
Niacinamide	0.00003663	0.00611	0.0000366
D-Pantothenic Acid•½Ca	0.000238	0.000477	0.000238
Pyridoxine•HCl	0.00006171	0.0000617	0.0000617
Riboflavin	0.0000376	0.000113	0.0000376
Thiamine•HCl	0.000337	0.000337	0.000337
Vitamin B-12	0.000407	0.000136	0.000136
<b>OTHER</b>			
Adenine•HCl	0.03088	0.00172	—
D-Glucose	1.081	1.441	1.8016
HEPES	6.6	7.149	—
Hypoxanthine	—	—	0.004083
Linoleic Acid	—	0.0000841	0.0000841
Phenol Red•Na	0.001242	0.001242	0.001242
Putrescine•2HCl	0.000161	0.000000161	0.000161
Pyruvic Acid•Na	0.055	0.055	0.11
Thioctic Acid	0.000206	0.00000206	0.000206
Thymidine	0.000727	0.0000727	—
<b>ADD</b>			
NaHCO <sub>3</sub>	1.176	N/A	1.18
Grams of powder required to prepare 1 L	17.7	17.7	11.0

**REFERENCES**

1. Boyce, S.T. and Ham, R.G., (1983). Calcium-Regulated Differentiation of Normal Human Epidermal Keratinocytes in Chemically Defined Clonal Culture and Serum-Free Serial Culture. *J. Invest. Dermatol*, 81, 33-40.
2. McKeenan, W.L. and Ham, R.G., (1976). Stimulation of Clonal Growth of Normal Fibroblasts with Substrata Coated with Basic Polymers. *J. Cell Biol.*, 71, 727-734.
3. Hamilton, W.G. and Ham, R.G., (1977). Clonal Growth of Chinese Hamster Ovary Cell Lines in Protein-Free Media. *In Vitro*, 13:9, 537-547.