M3:DC™ Chemically Defined and Clear Medium

Product Name: M3:DC™

Product Codes: M3DEC-100 and M3DEC-500

Product Use: Chemically defined and clear platform media for the M3™ media family, including M3Z™ media.

Features: GMP and clinical use quality; sterile; USP grade materials; xeno-free

INCELL CERTIFIED Chemically Defined No Animal-Derived Components

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General Description

M3:DC™ is a chemically defined, protein- and animal product-free solution of salts, sugars, amino acids, and buffers. M3:DC™ is NOT a complete cell culture growth medium since it does not contain growth factors, cytokines, blood source(s), or other supplements in the M3™/M3Z™ Media Family (Table 1).

Formulation and Packaging

M3:DC™ is the "platform media" formula of the M3™ media family including xeno-free M3Z™. It does not contain phenol red, so it is clear, but phenol red concentrate can be added to a concentration of 10 µg/mL. M3:DC™ derived complex media can have growth factor supplements from SMX-XF, human serum or human amniotic fluid without animal derived components so they are xeno-free (Table 1).

The M3™ media family was developed as "universal" media to support growth of human and other mammalian primary cultures, various fastidious stem cells and progenitors, tumor cells, and many other cell types (Tables 2 and 3). Supplemented media derived from M3:DC™ are clear and are highlighted in Table 1. M3Z™ media formulas are supplemented, designed for human use and based on M3™ media formulas. The M3Z™ media composition correlates with M3:Base media, but with no animal-derived components. Human male AB serum (huS) may be used as a M3Z™ supplement rather than fetal bovine serum (FBS), used in certain M3™ media.

Table 1. INCELL M3™ and M3Z™ Media Families of the M3™ "Super-Family"

M3™ Media Family							
Media Designation	**Product Code**	**Chem. Def'd**	**Phenol Red***	**SMX**	**Anti-biotics**	**Serum (% Source)**	**Amn. Fluid (% Source)**
M3:DC™	M3DEC	Yes	No	No	No	No	No
M3:BaseF™	M300F	No	Yes	Yes	No	No	No
M3:BaseA™	M300A	No	Yes	Yes	Yes	No	No
M3:10A™	M310A	No	Yes	Yes	Yes GCAm	Yes (10% FBS)	No
M3Z™ Media Family (No Phenol Red, Xeno-Free)							
Media Designation	**Product Code**	**Chem. Def'd**	**Phenol Red***	**SMX -XF**	**Anti-biotics**	**Serum or (% Source)**	**Amn. Fluid (% Source)**
M3Z:B™	MZ400	Yes	No	Yes	No	No	No
M3Z:H5 ™	MZ405	No	No	Yes	No	Yes (5% huS)	No
M3Z:H5A™	MZ405A	No	No	Yes	Yes, GC	Yes (5% huS)	No
M3Z:Y20™	MZ520	No	No	Yes	No	No	Yes (20% Amn. Fluid)

Legend. The M3™ Media Family includes M3™ (standard media) and M3Z™ (xeno-free; contains no animal-derived supplements; meets clinical use standards). The Table shows the attributes of the media manufactured and kept in stock. M3™ media uses M3:DC™ is the unsupplemented, chemically defined platform medium for the M3™ and M3Z™ families. M3:DC™ does not support cell culture growth alone but can be supplemented or used as control media for comparative studies. M3™ media for cell culture are highly enriched by multi-component supplement mix (SMX) which contains many growth factors, including some animal-derived growth components. In contrast, M3Z™ media are supplemented with SMX-XF which is xeno free. For serum ("S")- or amniotic fluid ("Y") -supplemented media, the (v/v) percent of serum or amniotic fluid are indicated by a number. As examples, the media designation "10" in M3:10A™ refers to 10% percent fetal bovine serum (FBS), "H5" refers to 5% human serum (type AB, male), and Y20 refers to 20% amniotic fluid. The media suffix designation "F" refers to "Free of antibiotics and serum". The "A" (Antibiotics) designation for any M3™ or M3Z™ media indicates that it contains gentamicin [G: 50 µg/mL] and clindamycin [C: 6.5 µg/mL]. The M3:10A™ media also contains Amphotericin B [Am; 2.5 µg/mL].*Sterile phenol red concentrate can be added to achieve 10 µg/mL.

Manufacturing

M3:DC™ is manufactured by sterile 0.22 µm filtration and packaging, using cGMP standards in an ISO Class 7 clean room and ISO Class 5 biosafety cabinet. Raw materials are pre-tested, and the final product is checked by quality specifications and acceptance criteria tested by USP standards for pH, osmolality, sterility (bacteria, fungi), mycoplasma and endotoxin prior to product release with a Certificate of Analysis.

Storage of M3:DC™ Media

M3:DC™ is stored refrigerated at 2°C to 8°C. The shelf-life is 24 months from the date of manufacture. Do not freeze.

CoA Specifications and Acceptance Criteria

Specifications	Acceptance Criteria
Visual	Clear, colorless to straw yellow
pH (USP <791>)	6.6 to 7.5
Osmolality (USP<785>)	280 to 340 mmol/kg
Sterility: SC (USP <71>)	No microbial growth
Sterility: fTG (USP <71>)	No microbial growth
Mycoplasma (USP <63>)	None detected
Endotoxin (USP <85>)	<0.5 EU/mL
Expiration	24 months from date of manufacture

M3:DC™
Chemically Defined Media

Use and Methods

Tables 2 and 3, respectively, describe extensive experience in use of INCELL supplemented M3™ media with many types of tissues and cells from humans and animals. A variety of methods to support cell monolayers and/or suspension cell growth have been used. Cell culture substrates have included standard plastics, complex biomatrices, meshes and engineered scaffolds.

Table 2. Human Tissues and Cells in M3™ Media for Oncology and Regenerative Medicine Clinical and Research Applications

Human Tissues	Primary Culture Cell Types or Cell Lines [Media]
Adipose (Fat)	Mesenchymal Stem Cells; Stromal vascular fraction regenerative cells [M3:10™]; adipose cells [M3:30™]
Bone Marrow; Bone, Cartilage, Adipocytes	Hematopoietic and mesenchymal stem cells; various types of renewable progenitor cells; endothelial cells; entire population [M3:20™]; cell subsets in other M3™ media; induced bone, cartilage or adipocyte outgrowth or induction of differentiation
Colon	Primary epithelial and/or mesenchymal support cells [M3:2™] [M3:10™] and INCELL Cell Line NCM460 [M3:10™]
Gastrointestinal	Primary epithelial cells and/or mesenchymal support cells; [M3:2™] [M3:5™] [M3:10™]
Kidney	Primary epithelial cells and/or mesenchymal support cells; [M3:2™] [M3:5™] [M3:10™]
Liver	Primary epithelial cells and/or mesenchymal support cells; [M3:2™] [M3:5™] [M3:10™]
Muscles (Heart; Peripheral; Smooth)	Pericytes; Mesenchymal or Stromal Stem Cells; regenerative cells [M3:10™]
Nucleus pulposus (NP) Intervertebral Disc	NP stem cells; annulus chondrocytes and mesenchymal stem cells; various types progenitor cells [M3:10™]; etc.
Pancreas	Pancreatic islet beta and acinar cells [M3:5™]; etc.
Peripheral or apheresis blood	Circulating or mesenchymal cells; endothelial cells [M3:20™]; subsets of cells in other M3™ media formulas
Placenta	Hematopoietic, endothelial and mesenchymal stem cells; trophoblasts; syncytiotrophoblasts; various renewable progenitor cells; [M3:10™]; others
Skin (adult; foreskin)	Epidermal keratinocytes co-cultures; Dermal Fibroblasts; Mesenchymal cells [M3:10™]; others
Tumors; various	Epithelial, mesenchymal, lymphoid; [M3:10™]; others
Umbilical Cord	Hematopoietic, endothelial and mesenchymal cells; various types of renewable progenitor cells; [M3:10™]; etc.

Table 3. M3™ Complete, Supplemented Media Have Been Used for Animal Tissues/Cells Cultured for Biomedical Research

Cells and Tissues Derived from Adult, Newborn and/or Fetal Sources	
Animal Tissues	**Species & Cultured Cell Types/ Cell Lines [Media]**
Adipose (Fat)	Rat, mouse, hamster, rabbit; Stromal vascular fraction regenerative MSC cells [M3:10™]; adipose cells [M3:30™]*
Bone Marrow	Rat, mouse, hamster, rabbit; Hematopoietic and mesenchymal stem cells; various types of renewable progenitor cells; Endothelial cells; entire population [M3:20™]; subsets of cells in other M3™ media
Brain & Neural (Spinal)	Rat, mouse: Progenitors; induced growth and differentiation; [M3:5™] [M3:10™]
Colon; Gastrointestinal	Rat, mouse, hamster; Primary epithelial or mesenchymal support cells [M3:2™]; [M3:5™]; [M3:10™] or complex tissues in organ-like cultures
Kidney	Rat, mouse, hamster; Primary epithelial cells and/or mesenchymal support cells; [M3:2™]; [M3:5™]; [M3:10™]
Liver	Rat, mouse, hamster; Primary epithelial cells and/or mesenchymal support cells; [M3:2™]; [M3:5™]; [M3:10™]
Muscles (Peripheral; Heart; Smooth)	Rat, mouse, hamster, rabbit; Pericytes; Mesenchymal or Stromal Stem Cells; regenerative cells [M3:10™]
Pancreas; Other Neuroendocrine	Rat, mouse, hamster; pancreatic islet beta and acinar cells [M3:5™]; other organs (e.g., adrenal)
Peripheral or apheresis blood	Rat, mouse, hamster, rabbit; Circulating or mesenchymal cells; endothelial cells [M3:20™]
Skin (adult; newborn)	Epidermal keratinocytes co-cultures; Dermal Fibroblasts; Mesenchymal cells [M3:10™]; others
Tumors; various	Rat, mouse, hamster; epithelial, mesenchymal, lymphoid; [M3:10™]; etc.

Legend to Tables 2 and 3. Supplemented M3™ media formulations may contain various percentages of FBS added to M3Base™. Those media are designated as M3:{% serum}. As an example, M3:10™ contains 10% v/v FBS. Some media are kept in stock, while others are special manufactured or made by the customers by adding the FBS to the media.

Master Files Applications Note

M3:DC™, as part of the M3 Media Family, is in FDA Drug and Device Master Files but have not been tested by INCELL for any specific diagnostic or therapeutic use. To request use of a Master File call, FAX, or email to info@incell.com.

Order: Contact INCELL

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Technical Assistance

The scientists at INCELL are available to discuss the media or special needs of your cells and to assist you in your cell culture applications. Call 1-800-364-1765 or e-mail info@incell.com.