CytoNiche
Architect for Cells

3D TableTrix® Microcarriers

Porous & Dissolvable Pharmaceutical-grade Microcarriers
for Adherent Cell Mass Manufacturing

Architect for Cells
—— Expert in 3D manufacturing of high quality cells

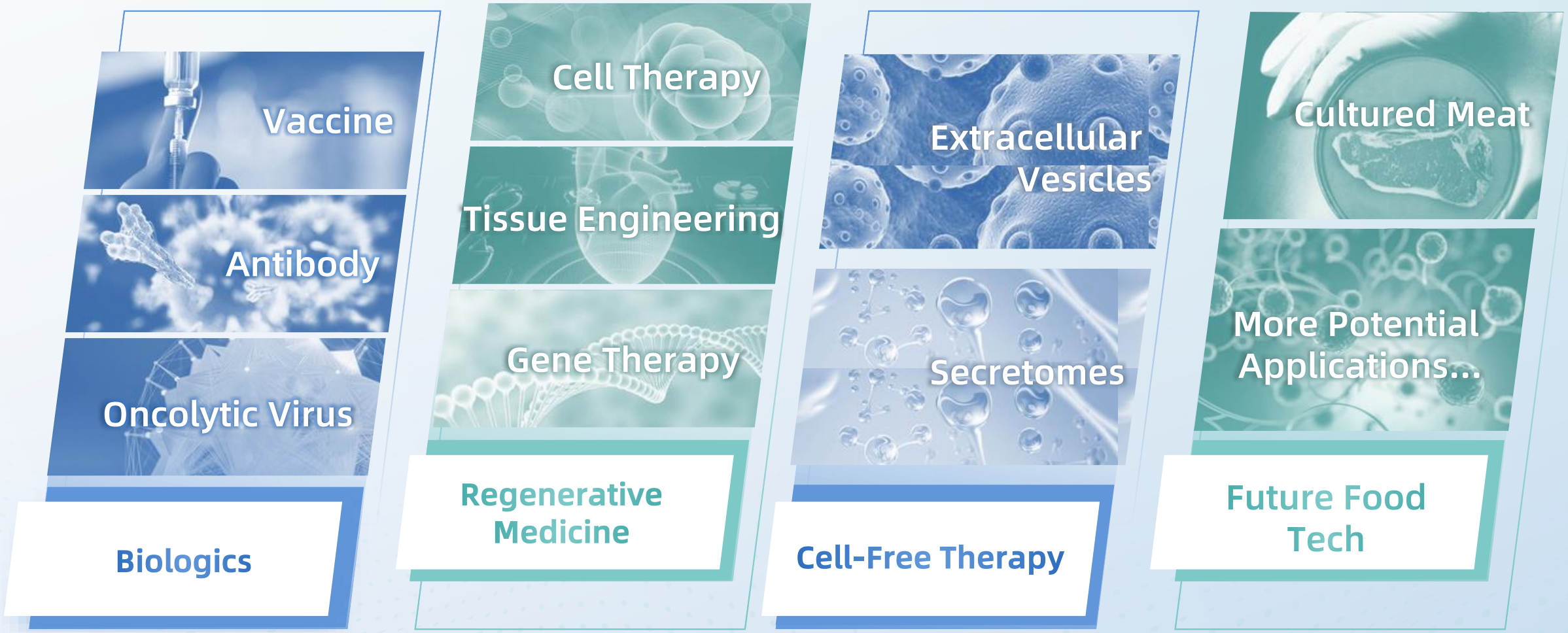
www.cytoniche.com

CytoNiche
Architects for Cells

Cell Mass Manufacturing

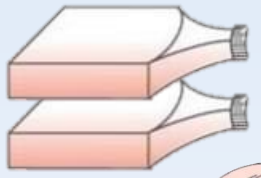
Who Needs It & How It is Done

Biotech Sectors in Need of Cell Mass Manufacturing

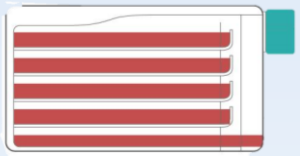


Options for Cell Manufacturing

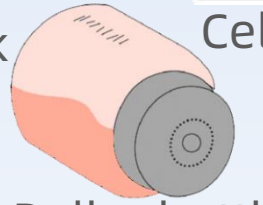
2D Culture



T-flask



Cell factory



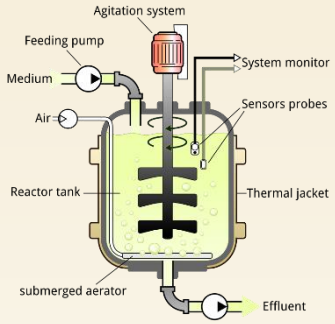
Roller bottles

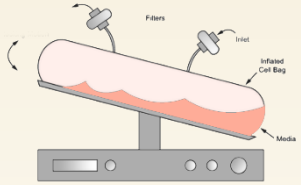
Common cell types:
Most adherent cells

Industrial applications:
- Stem cell therapy
- Diploid cells for vaccine production
- Virus packaging with HEK293T
- … others that current suspension or microcarriers cannot support

Suspension Culture

Shaker flask



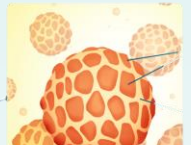


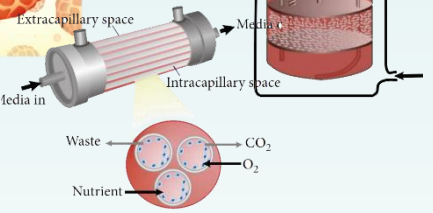
Bioreactors

Common cell types:
Adapted suspension mammalian cells (CHO, 293F, MDCK)
Insect cells
Immune cells (T cells, NK, etc)

Industrial applications:
Biologics/Vaccines & proteins
Gene therapy/Virus packaging
Immune cell therapy

Microcarriers & Fibers



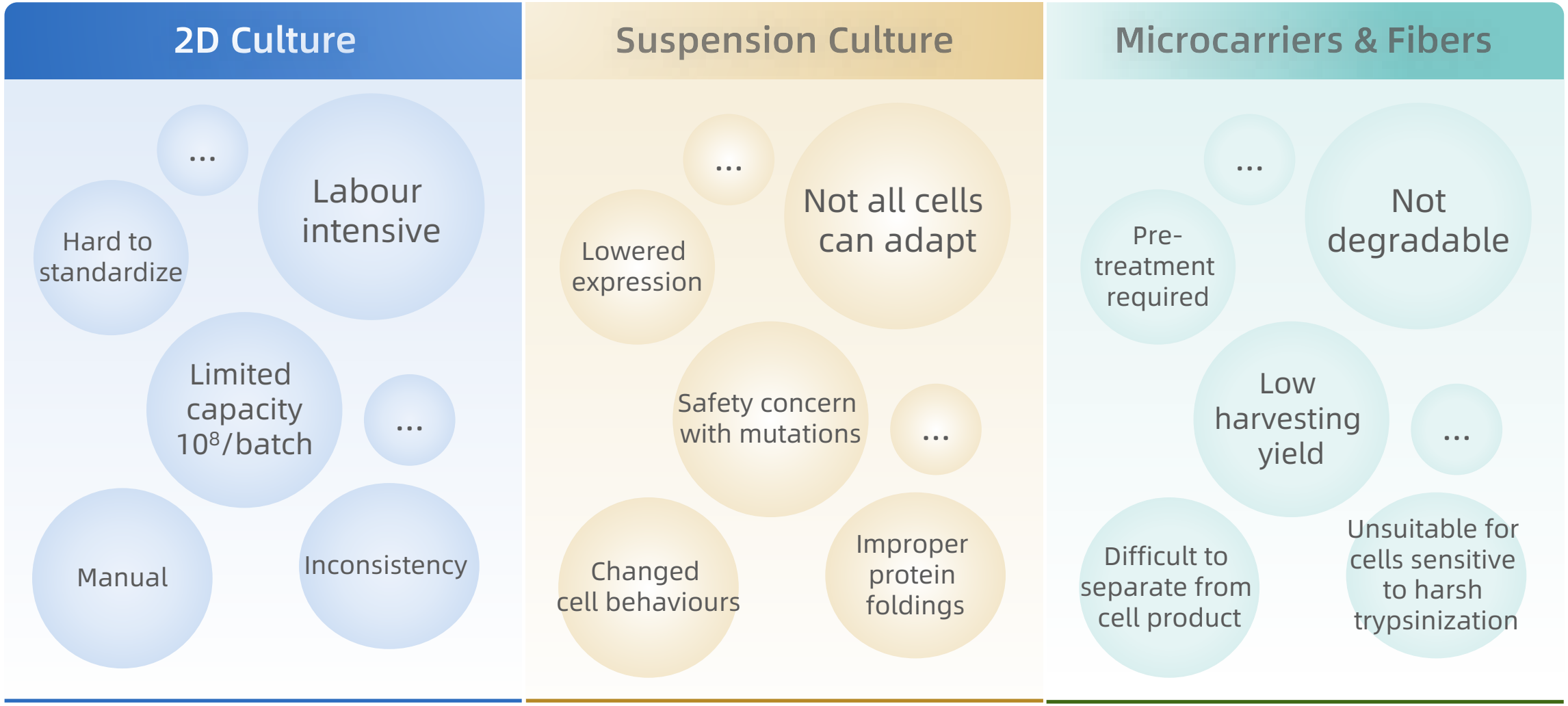


Microcarriers
Hollow Fibers
Fixed-bed Bioreactors

Common cell types:
Adherent mammalian cells
(293, adherent CHO, VERO, MARC145, immortalized hepatic cells, etc.)

Industrial applications:
Biologics/Vaccines & proteins
Gene therapy/Virus packaging
Exosomes

Limitations



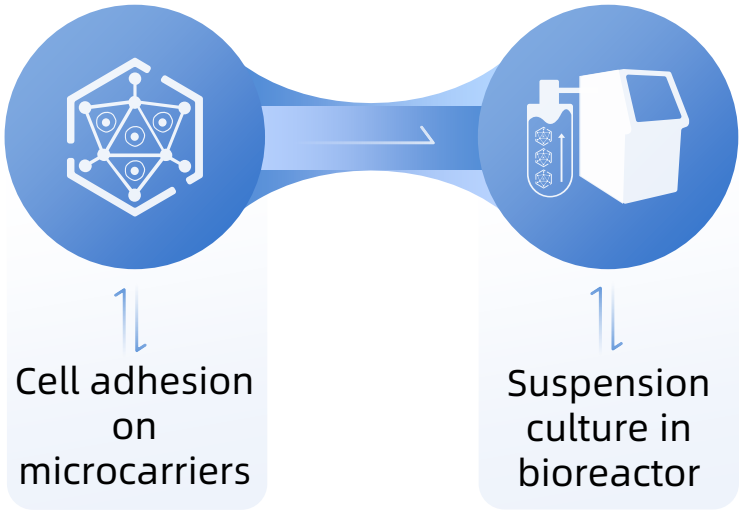
CytoNiche
Architects for Cells

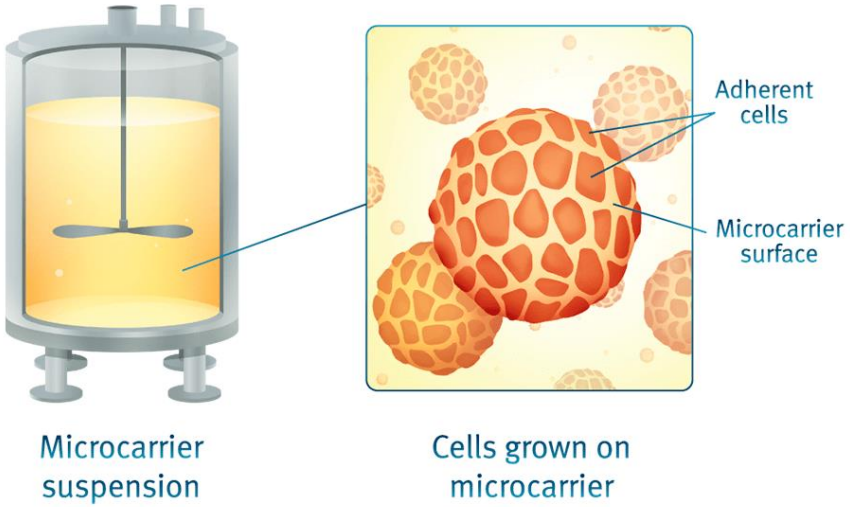
3D TableTrix® Microcarriers

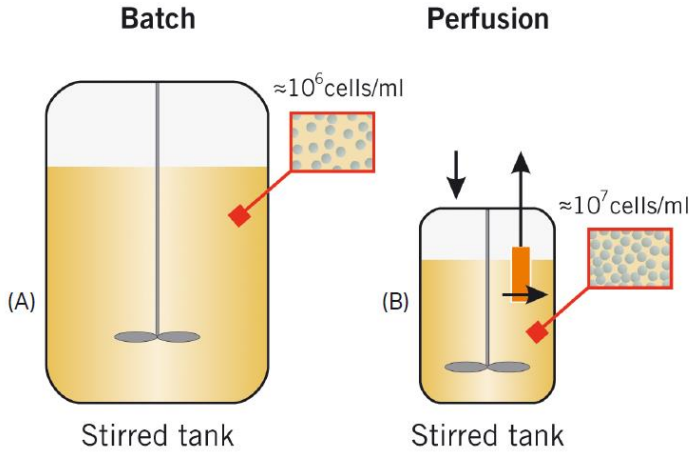
Dissolvable porous microcarriers

What is Microcarrier

- A micron-size support matrix that allows for the growth of adherent cells in bioreactors
- Usually exist as 100–300 μm polymeric beads

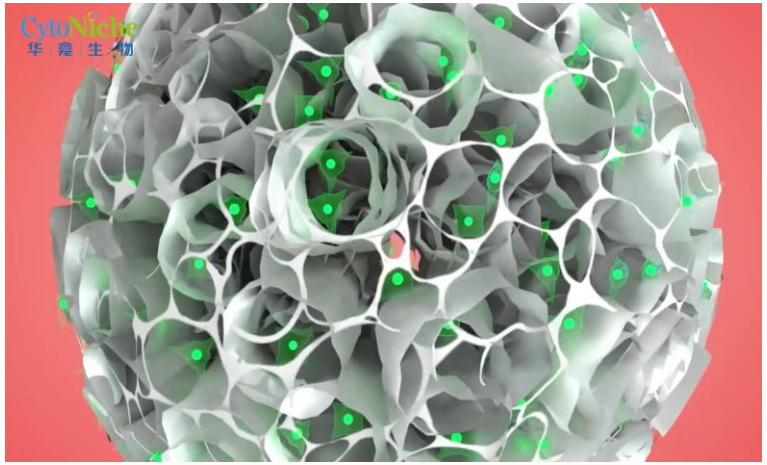


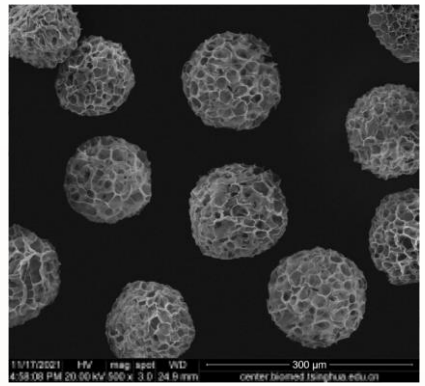


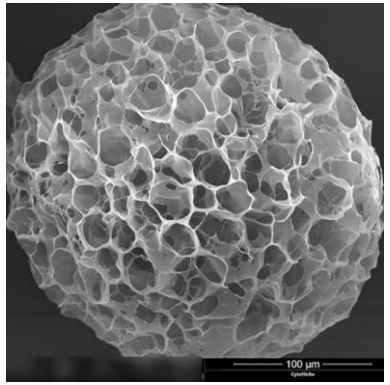


3D TableTrix® Microcarriers: Unique Features

Dissolvable
>99% recovery of cells







High Porosity
Up to 9000 cm^2/g
Yield 10^8~10^9 cells

Pharmaceutical Grade
Produced under GMP
Registered with U.S. DMF

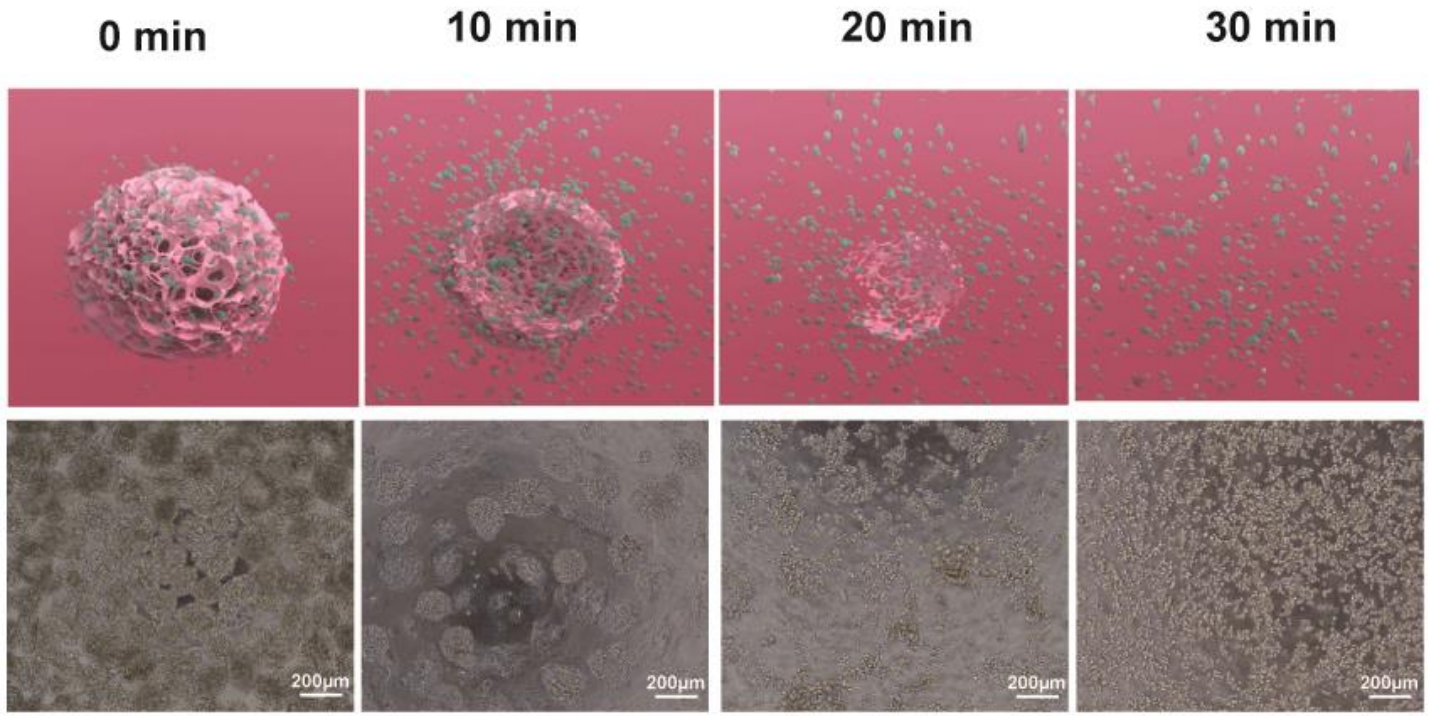
Ready-to-use
Sterile, weight-defined*

*Applicable to some products only

Unique Features: Dissolvable

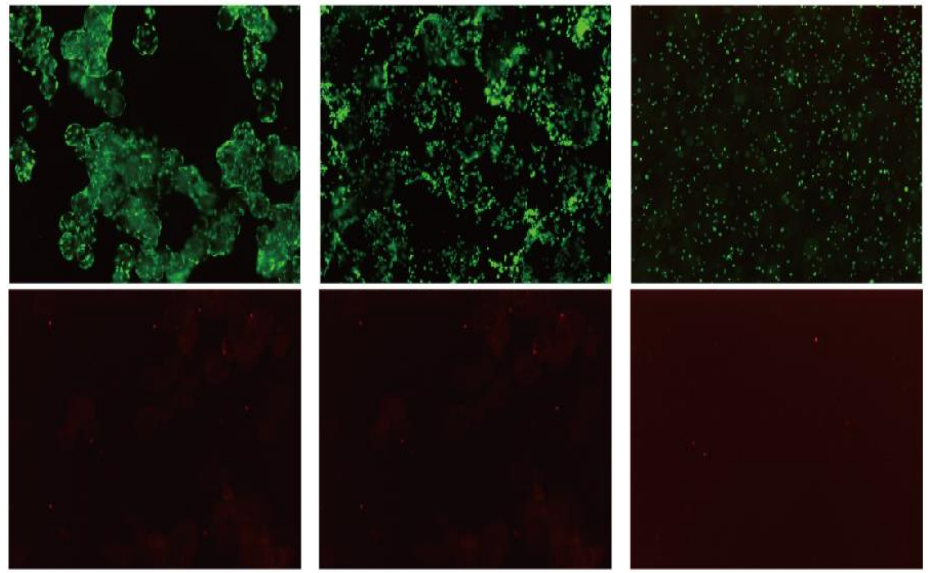
Dissolvable
>99% recovery of cells

Complete cell recovery from microcarriers
by dissolving with 3D FloTrix® Digest



Performed at 37°C, pH 7.0-7.4

Cells remain viable during process

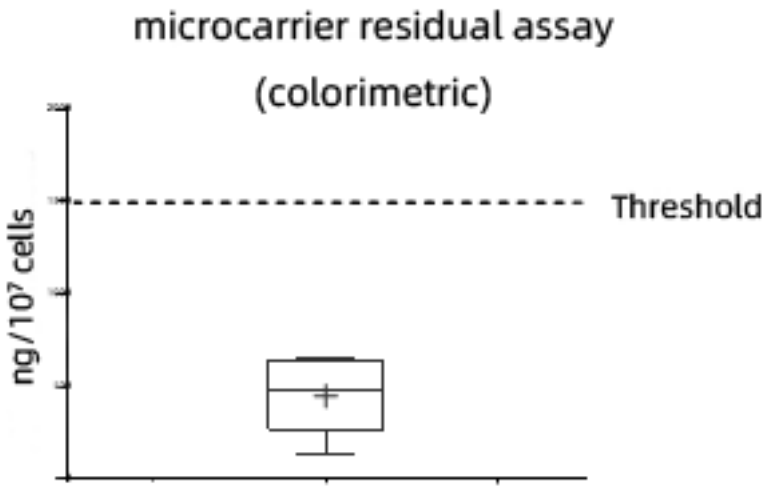


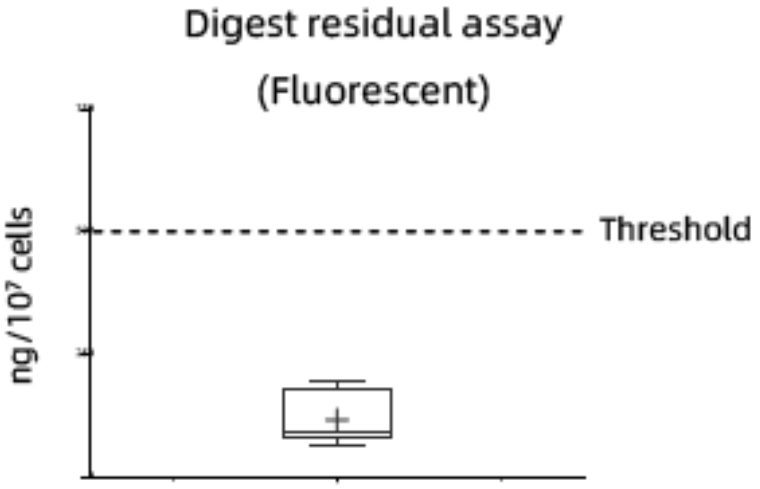
LIVE/DEAD

Unique Features: Dissolvable

Low residual content in end products
—— remove simply by centrifuge

Accredited by National Institutes for Food & Drug Control (China)





No toxicity was observed in animals injected with 1X, 10X or 100X concentrations of residual content thresholds

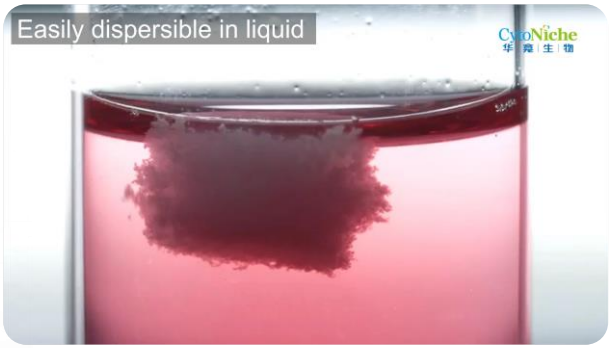
Dissolvable: Why is It Important?

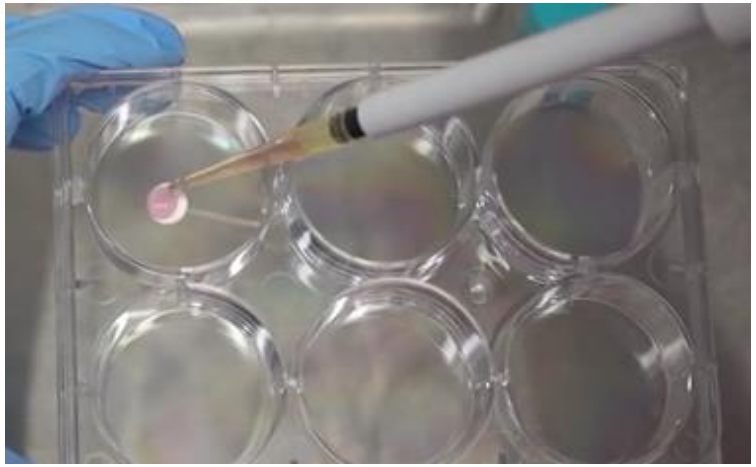
- Cells are THE "**living**" **end-product** of cell therapy
  - Downstream processing methods for live cells are limited
  - Extensive purification cannot be performed
  - Need to keep residuals from starting materials used in the manufacturing process low to ensure safety
- Good **quality seed cells** ensure efficient biologic production
  - Large quantity of cells needed for the final stage of the upstream process (i.e. virus production, protein production) in the largest bioreactor (up to 2000 L bioreactors)
  - Harsh detachment methods used in conventional undegradable microcarriers are not suitable for delicate cells, i.e. diploid cells
  - Biomimetic environment to trigger better virus production

Unique Feature: Dispersible Tablets

Ready-to-use
Sterile, weight-defined*

- **Ready to use**
  - Pre-sterilized
  - Closed system packaging for bioreactors (* powder only)
- **Designed for lab-scale process**
  - Defined weight per tablet (20 mg)
  - No hassle of weighing powder
  - Static culture in 6-well plates (one tablet/well)
  - Dynamic culture in spinner flasks or with microSPIN* system
- **Dispersible**
  - **NO** adhesive or additional chemical substances
  - **NO** disintegrants
  - Tablet disperses into individual microcarriers when in contact with liquid





* Bioreactor designed with 6-well plates with built-in impellers, available from CytoNiche

Unique Feature: Safe for cellular drugs

Pharmaceutical Grade
Registered with U.S. DMF

Approved by FDA

DMF # 35481

One and only microcarriers listed by FDA under DMF filings

Pharmaceutical Excipients Recognized by the Center for Drug Evaluation (China)

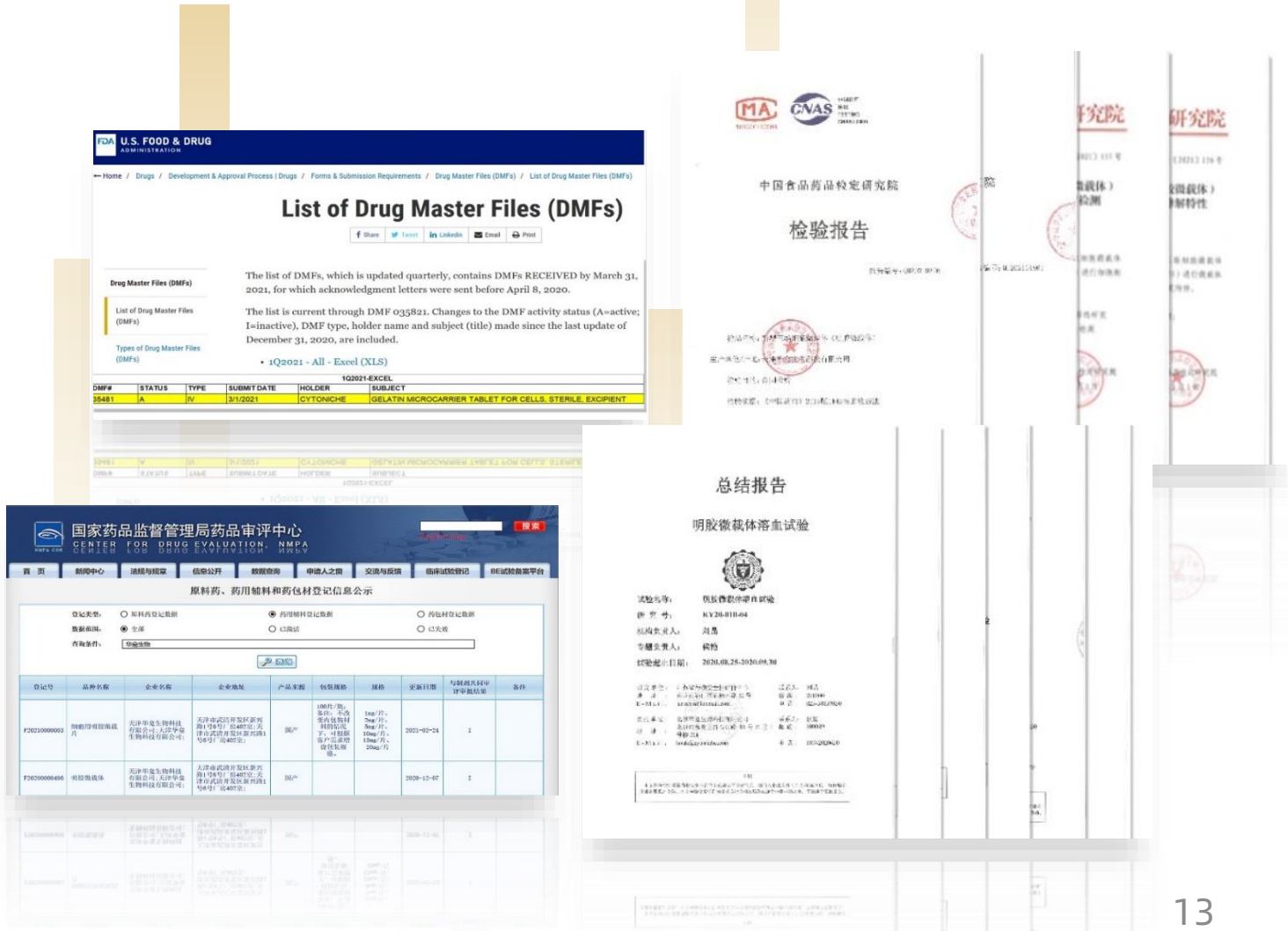
Registration No.

#F20200000496

#F20210000003

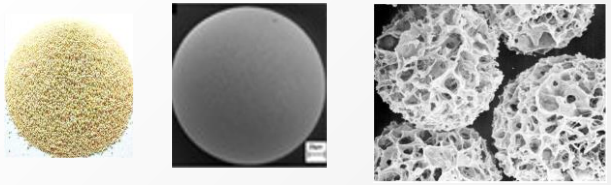
Quality and Safety Report

- ✓ Biocompatibility
- ✓ Stability
- ✓ Cytotoxicity
- ✓ Acute Toxicity (*in vivo*)
- ✓ Allergic reaction
- ✓ Immunogenicity
- ✓ Hemolysis test
- ✓ Pyrogenic reaction
- ✓ ...



3D TableTrix® Microcarriers: Comparison

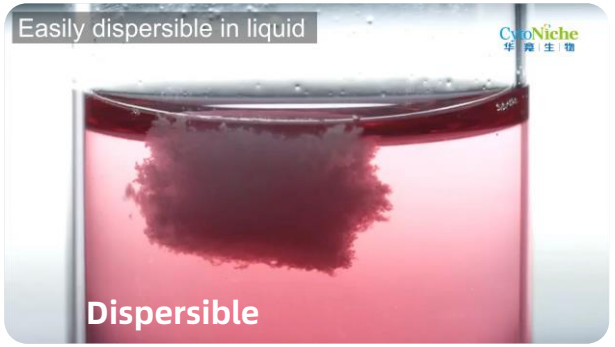
Conventional Microcarriers

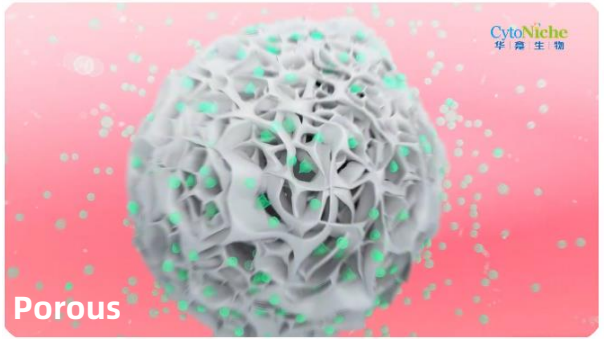


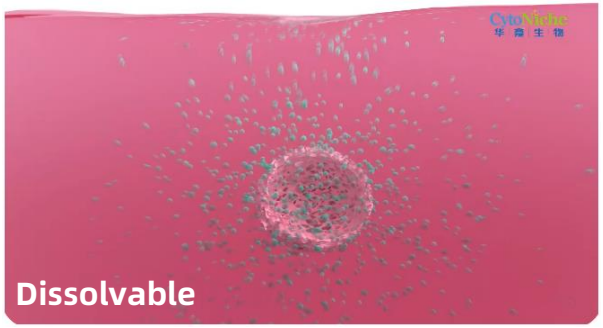
- Non-degradable
  - Harsh harvesting condition
  - Extra separation protocol
- Pre-treatment needed
  - Powder in bulk only
  - May need bead hydration
- Solid sphere or macroporous
- Synthetic polymers

3D TableTrix® Microcarriers

- ✓ **Fully dissolvable**
  - ✓ Gentle harvesting
  - ✓ No extra separation
- ✓ **Ready-to-use***
  - ✓ Weight-defined tablets
  - ✓ Dispersible
- ✓ **Macroporous**
- ✓ **Biomimetic protein**







*Choose from:
- Ready-to-use weight-defined dispersible tablet for lab scale
- Powder in bulk for mass production (closed system packaging)

3D TableTrix® Microcarriers: Comparison

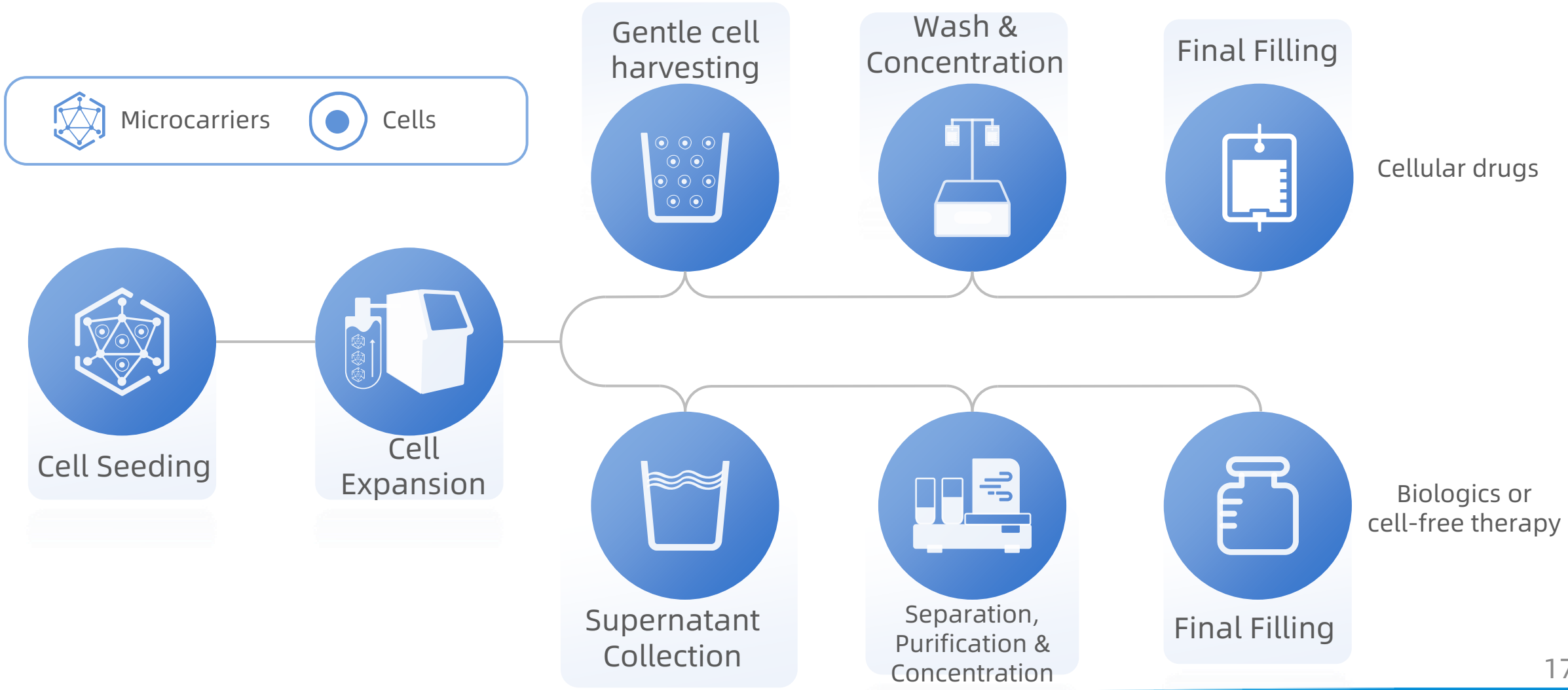
Product / Feature	Cytodex-1	Cytodex-3	SoloHill	Cytopore1	Cytopore2	3D TableTrix W01	3D TableTrix G02	3D TableTrix V01
Material	Dextran	Dextran	Polystyrene	Cellulose	Cellulose	Denatured collagen	Denatured collagen	Denatured collagen
Structure	Solid Sphere	Solid Sphere	Solid Sphere	Porous Sphere	Porous Sphere	Porous Sphere	Porous Sphere	Porous Sphere
Size（μm）	147-248	133-215	90-150/125-212	200-270	200-270	100-200	120-180	80-200
Pore（μm）	/	/	/	30	30	30	30	30
Surface Area (cm^2/g)	4400	2700	360/480	~11000	~11000	>6000	>6000	>6000
Surface Modification	DEAE positive charge	Gelatin	RGD/Positive charge/Gelatin	DEAE positive charge	DEAE high positive charge	N/A	N/A	N/A
Packaging	Powder	Powder	Powder	Powder	Powder	Sterile Tablet/Powder	Sterile Tablet/Powder	Powder
Harvest Efficiency	Poor	Poor	Poor	Not possible	Not possible	High	High	High
Degradability	NO	NO	NO	NO	NO	YES	YES	YES
Application Field	Biologics	Biologics	Biologics	Biologics	Biologics	Cell & Gene Therapy Biologics	Cell & Gene Therapy Biologics	Biologics

3D TableTrix® Microcarriers: Applications

Product / Feature	3D TableTrix W01	3D TableTrix G02	3D TableTrix V01
Material	Denatured collagen	Denatured collagen	Denatured collagen
Size（μm）	100-200	120-180	80-200
Packaging	Tablet/Powder	Tablet/Powder	Powder
Pre-sterilized	Yes	Yes	No
Suitable cell types	Mesenchymal stem cells	VERO MDCK 2BS/MRC-5 Fish primary cells	HEK293T Adherent CHO
Suitable application	Working cell bank expansion Final cell production Exosome production	Seed cell expansion Human/Animal vaccine production Oncolytic virus production	Lenti-virus production Adeno-associated virus production Protein expression

Cell Mass Manufacturing Process with 3D TableTrix®

Use of 3D TableTrix® microcarriers for adherent cell manufacturing



CytoNiche Architects for Cells

Cell Types We Have Worked With

Cell Therapy

- hUC-MSC
- Placenta MSC
- Adipose-derived MSC
- Amniotic membrane MSC
- Dental Pulp MSC
- ESC-derived MSC
- Bone Marrow MSC
- iPSC-derived MSC

Vaccines

- MRC-5
- Vero
- 2BS
- PK-15
- Diploid cells
- MARC-145
- MDCK
- Fish cells

Virus Packaging

HEK293T cell line

Artificial Liver

Immortalized Human Hepatocytes

Other Cell Types

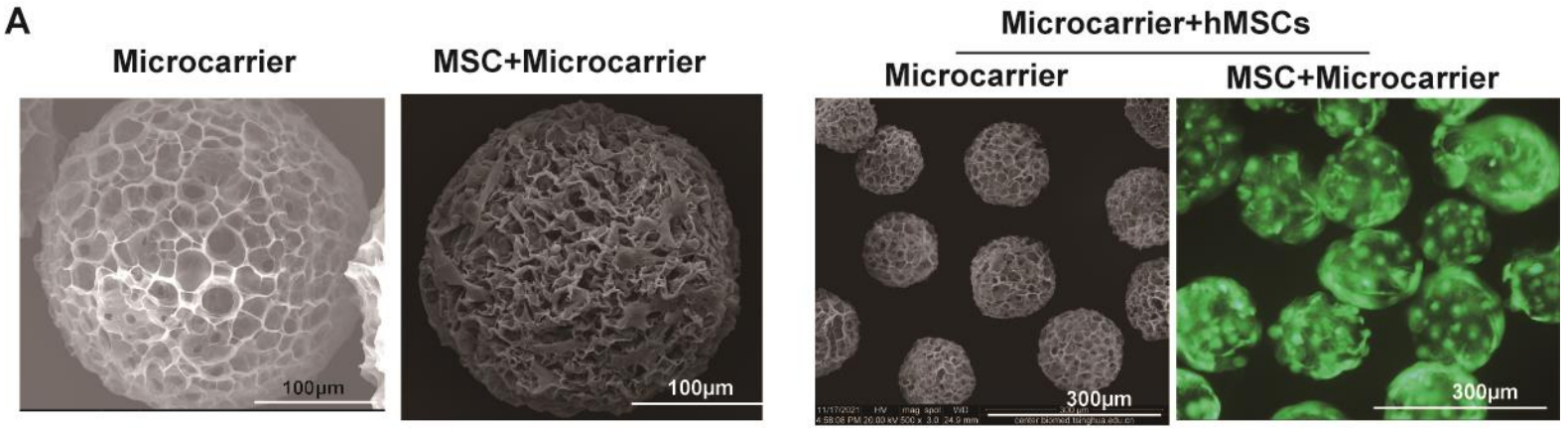
- hAEC (Human amniotic epithelial stem cells)
- HDF Human Dermal Fibroblasts
- UPC human kidney adult stem cells
- 3T3 Cell Lines
- Nasal mucosal stem cells
- Neurons
- HUVEC

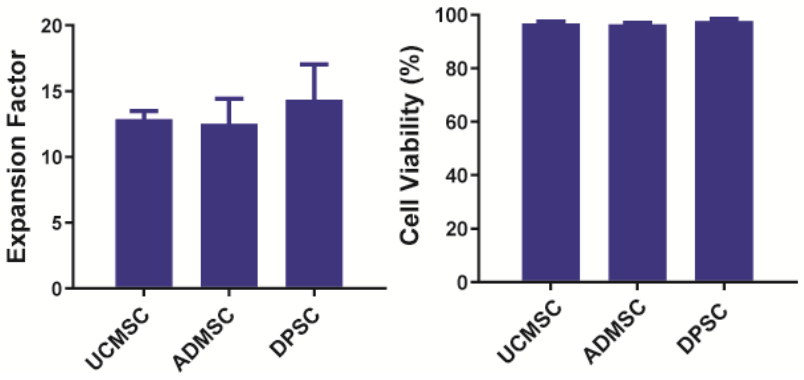
Cultured Meet

Porcine Muscle Stem Cells
Porcine Adipose Stem Cells

CytoNiche Architects for Cells

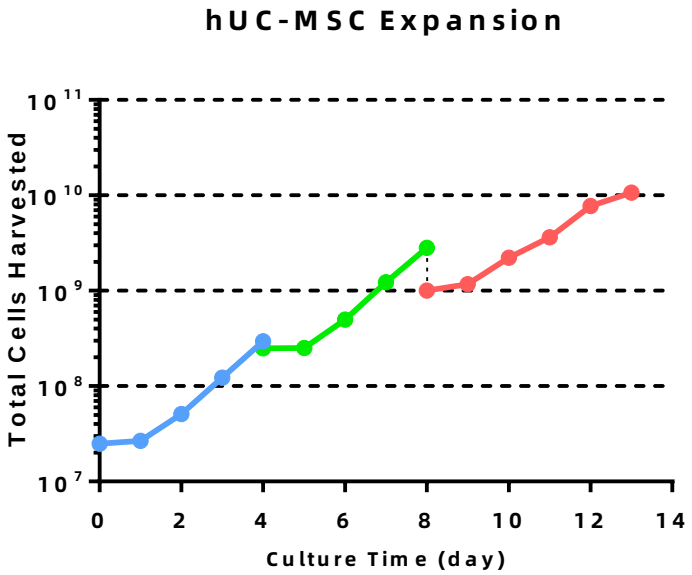
Performance Data: MSCs with W01 microcarriers



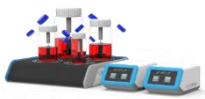


Serum-free medium, 4-5 days culture

Data from Journal of Tissue Engineering and Regenerative Medicine, 2022, 16(10), 934-944.



3D FloTrix® miniSPIN



Phase 1

3D FloTrix® vivaSPIN (5L)

Phase 2

3D FloTrix® vivaSPIN (15L)

Phase 3

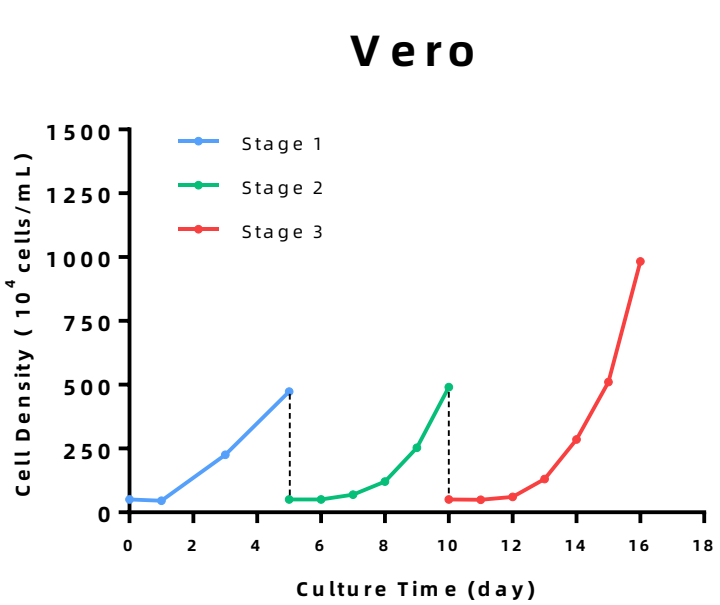
3D FloTrix® vivaPREP PLUS

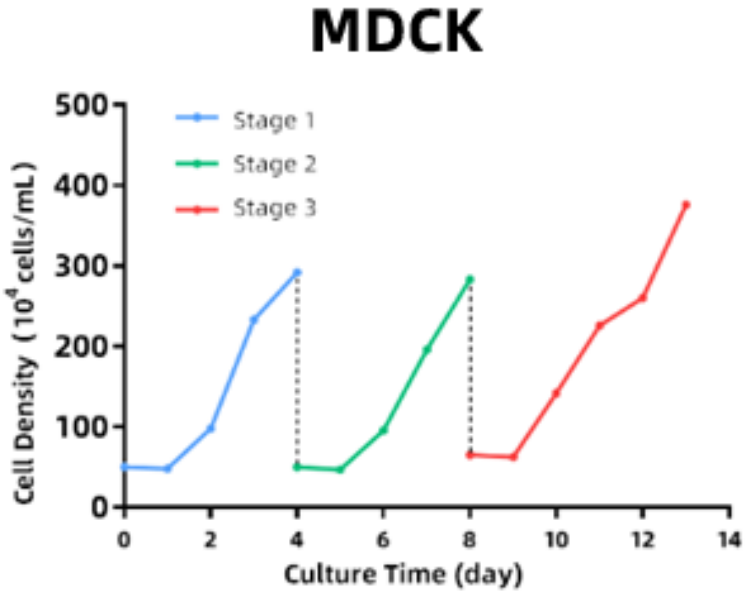
Wash & Concentrate

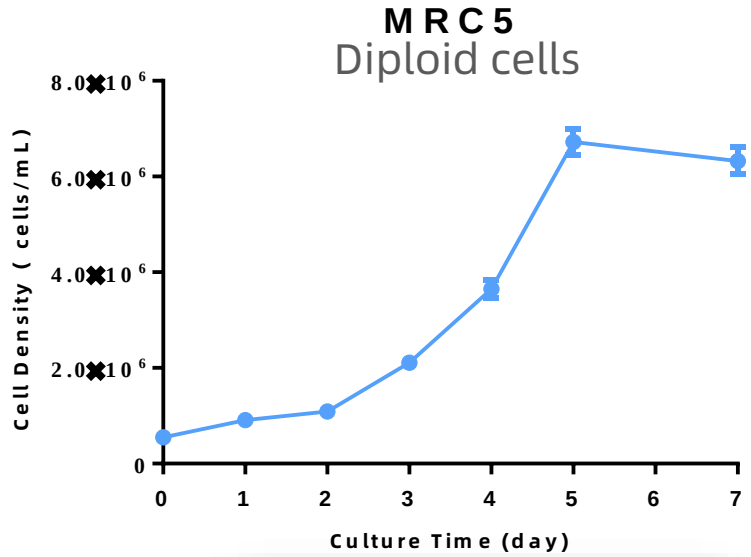
Check MSC deck for more details on cell quality & performance comparison with other microcarriers

CytoNiche Architects for Cells

Performance Data: V01 microcarriers





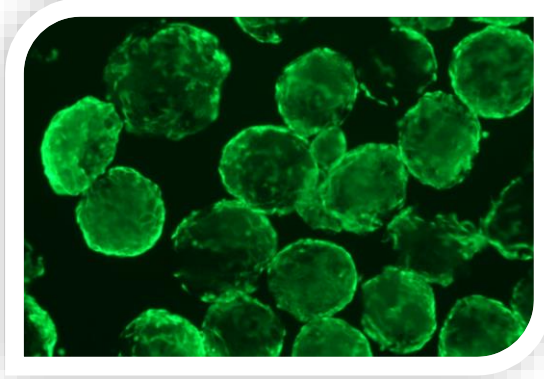


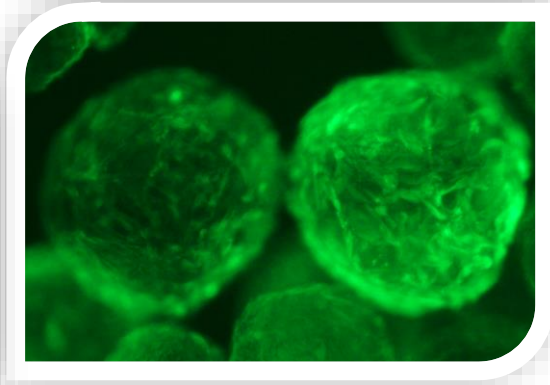
Performance of V01 Microcarrier

10^7 cells/mL is achievable

Performance validated in 300 L bioreactor

Bead-to-bead transfer for passaging

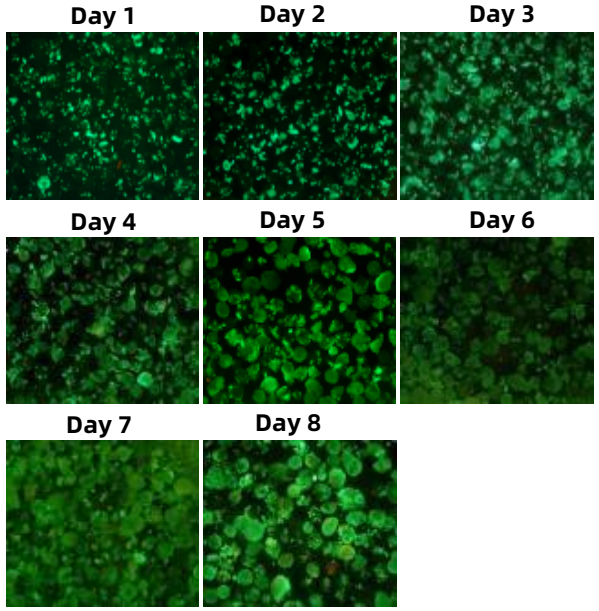


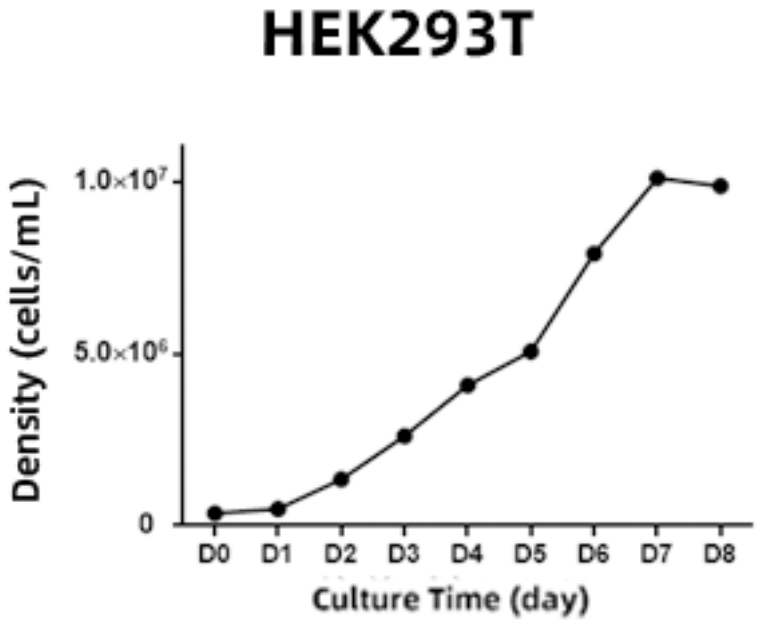


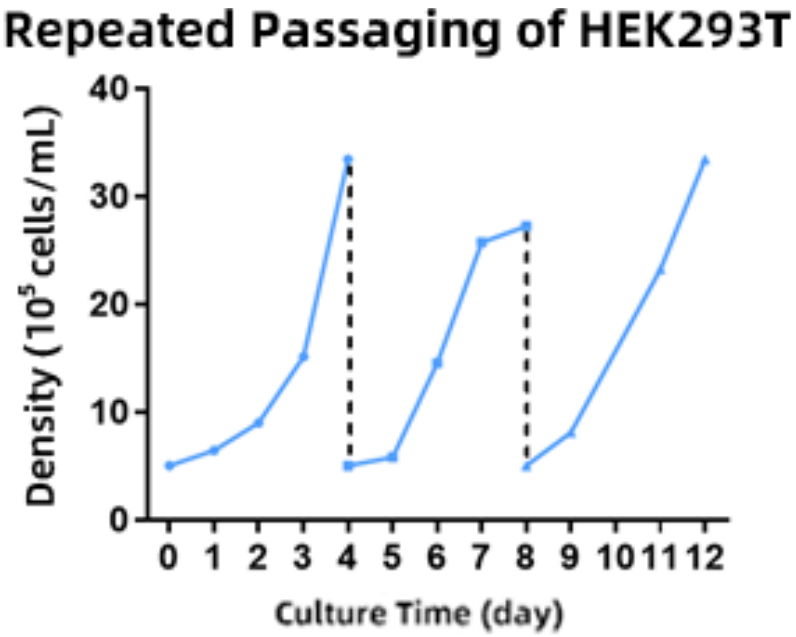
Check Vaccine & Virus deck for more details on more vaccine virus titer & performance comparison with other microcarriers & 2D

CytoNiche Architects for Cells

Performance Data: HEK293T on G02 microcarriers







Performance of G02 Microcarrier

10^7 cells/mL of HEK293T is achievable

Density comparable to suspension culture

Bead-to-bead transfer for passaging

Check HEK293T deck for more details on more plasmid transfection efficiency, lenti-virus & adeno-associated virus titer, & cell culture performance comparison with other microcarriers

Performance Data: various cell types

Cells	Culture Medium	Culture time (days)	Expansion Factor	Cell Viability
MSCs from various tissue sources	Serum-free	4-6	8-14	≥95.7%
Porcine muscle stem cells	Serum-free	7	16.52	95.1%
Human Hepatic Cells	Serum-free	7	2.8	90.3%
HUVEC	With serum	10	35	95.5%
3T3	With serum	6	24.55	92.6%
293T	With serum	4	9.3	94.0%
PK15	With serum	3	5.4	90.7%
Vero	With serum	4	8.2	90.4%
MRC-5	With serum	4	5.0	97.47%

Holistic Solution for Cell Mass Production

CytoNiche offers various products & extensive technical support to accelerate your cell manufacturing process

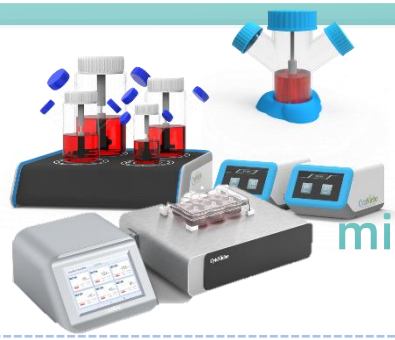
Reagents and Consumables

3D TableTrix®
Microcarriers

3D FloTrix®
Digest for microcarriers

3D FloTrix®
MSC SF Medium

Bioreactors



3D FloTrix®
micro/miniSPIN
Bioreactors

3D FloTrix®
vivaSPIN
Stir-tank
Bioreactors

Single-use

3D FloTrix®
vivaROCK
Bioreactors

Downstream Processing

3D FloTrix®
vivaPREP/vP PLUS
cell processing system

3D FloTrix®
vivaEXO
Exosome Collection System

Holistic Solution for Cell Mass Production

New products are coming Sep 2023 to bring cell mass production at GMP-standard a step further.

Microcarriers

Animal Component Free Dissolvable Microcarriers

Bioreactors

3D FloTrix® miniSPIN FLEX

Independent control for each channel
Ultra slim to fit 2 sets into an incubator
with disposable spinner flasks

Downstream Processing

3D FloTrix® vivaPACK
cell formulation
& filling system
(cryobag)/(cryovial)

Publications

- **Dispersible and dissolvable porous microcarrier tablets enable efficient large-scale human mesenchymal stem cell expansion (2020). *Tissue Engineering Part C: Methods*, *26*(5), 263-275.**

- **GMP-grade microcarrier and automated closed industrial scale cell production platform for culture of MSCs (2022). *Journal of Tissue Engineering and Regenerative Medicine*, *16*(10), 934-944.**

- Engineering 3D functional tissue constructs using self-assembling cell-laden microniches (2020). *Acta biomaterialia*, *114*, 170-182.
- Intra-articular injection of cell-laden 3D microcryogels empower low-dose cell therapy for osteoarthritis in a rat model (2020). *Cell transplantation*, *29*, 0963689720932142.
- A low dose cell therapy system for treating osteoarthritis: In vivo study and in vitro mechanistic investigations (2022). *Bioactive Materials*, *7*, 478-490.
- Exendin-4 gene modification and microscaffold encapsulation promote self-persistence and antidiabetic activity of MSCs (2021). *Science Advances*, *7*(27), eabi4379.
- Cell-subpopulation alteration and FGF7 activation regulate the function of tendon stem/progenitor cells in 3D microenvironment revealed by single-cell analysis (2022). *Biomaterials*, *280*, 121238.
- Efficient endothelial and smooth muscle cell differentiation from human pluripotent stem cells through a simplified insulin-free culture system (2021). *Biomaterials*, *271*, 120713.
- Engineered meatballs via scalable skeletal muscle cell expansion and modular micro-tissue assembly using porous gelatin micro-carriers (2022). *Biomaterials*, *287*, 121615.
- Exosomes derived from three-dimensional cultured human umbilical cord mesenchymal stem cells ameliorate pulmonary fibrosis in a mouse silicosis model (2020). *Stem Cell Research & Therapy*, *11*(1), 1-12.
- Exosomal let-7i-5p from three-dimensional cultured human umbilical cord mesenchymal stem cells inhibits fibroblast activation in silicosis through targeting TGFBR1 (2022). *Ecotoxicology and Environmental Safety*, *233*, 113302.

CytoNiche
Architect for Cells

Empowering cell and gene therapy with intelligent 3D cell mass manufacturing technology to benefit more patients

Architect for Cells

—— Expert in 3D manufacturing of high quality cells

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Architects for Cells

Launching a new era for industrialising cell manufacturing

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