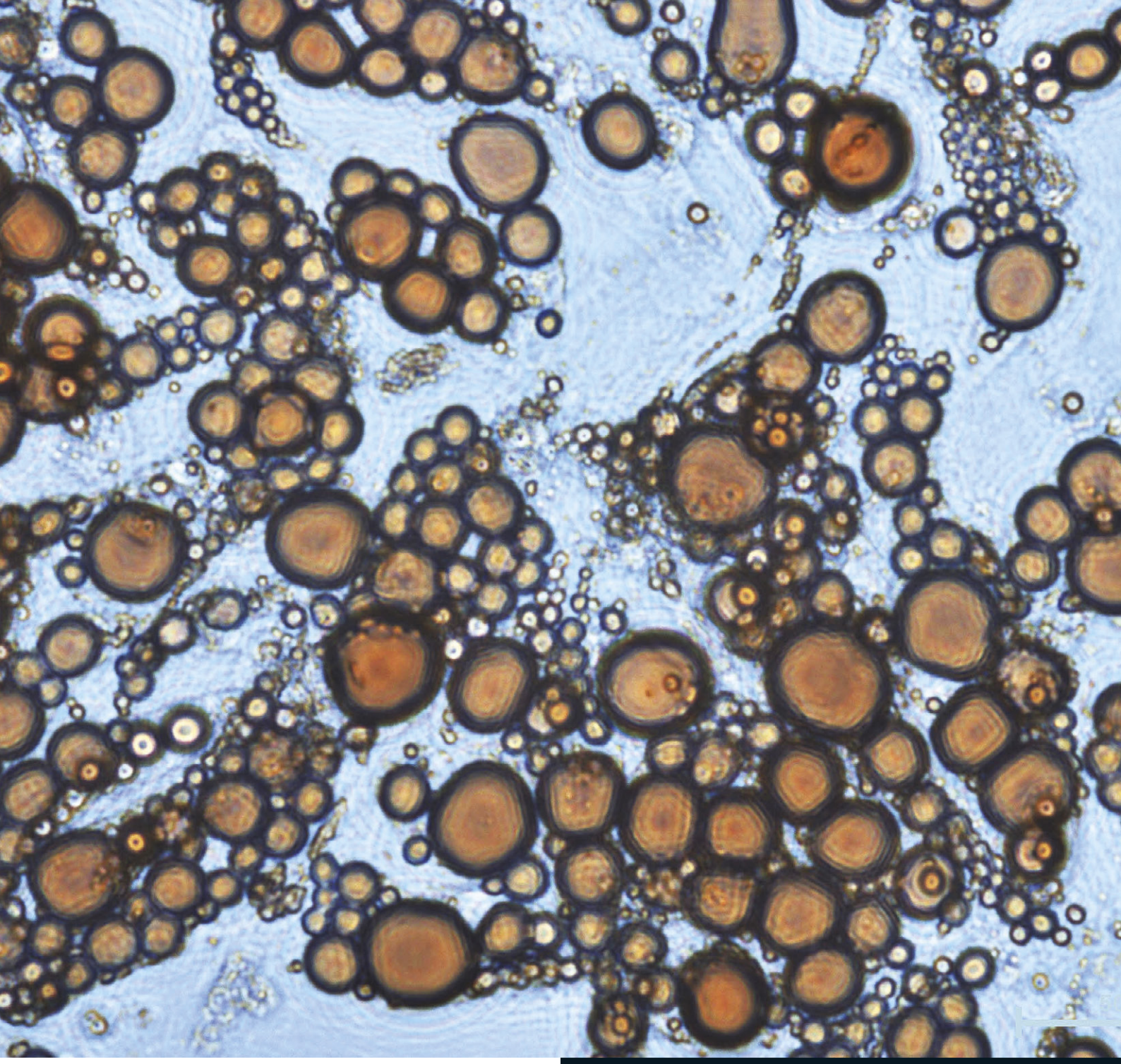


Human Mesenchymal Stromal Cells

Serum-Free, Xeno-Free Systems for the Culture and Differentiation of Human Mesenchymal Stromal Cells

Simplifying Progress

SARTORIUS

Seamless Transition From Research to the Clinic

Human Mesenchymal Stromal Cells (hMSC) are multipotent adult stem cells present in a variety of tissue niches in the human body such as Adipose Tissue (AT), Bone Marrow (BM), Placenta (PL) and Wharton's Jelly (WJ). hMSC have advantages over other stem cell types due to the broad variety of their tissue sources, for being immunoprivileged, and for their ability to specifically migrate to tumors and wounds **in vivo**.

Due to these traits hMSCs have become desirable tools in tissue engineering and cell therapy. In most clinical applications hMSCs are expanded **in vivo** before use. The quality of the culture medium and its performance are particularly crucial with regard to therapeutic applications, since hMSC properties can be significantly affected by medium components and culture conditions. A defined serum-free, xeno-free culture system optimized for hMSC isolation and expansion greatly facilitates the development of robust, clinically acceptable culture processes, reproducibility and generating quality-assured cells.

Sartorius offers a novel serum-free (SF) and xeno-free (XF) culture system, which includes specially developed solutions for the attachment, dissociation and cryopreservation, as well as MSC NutriStem® media, which enable long-term growth of hMSCs from various sources while retaining self-renewal and multi-lineage differentiation potential.

In addition to the culture system, Sartorius offers serum-free, xeno-free media for the direct differentiation of hMSCs from various sources into adipocytes, chondrocytes and osteocytes. The differentiation media contain all the growth factors and supplements necessary for the directed differentiation of hMSCs.

hMSC culture system	Cell growth	Long-term cell characteristics	Ease of use	Clinical compliance	Lot consistency
MSC NutriStem® XF + Attachment Solution	+++	+++++	++	++++	++++
MSC NutriStem® XF + NutriCoat™	+++	+++++	+++	+++++	+++++
MSC NutriStem® XF + 2% PLTGold®	+++++	+++++	++++	+++	+++
MSC NutriStem® XF + CellBIND®	+++	+++++	+++++	+++++	+++++
MSC NutriStem® Basal Medium + 5% PLTGold®	++++	+++	+++++	+++	+++
MEM-a + 5% PLTGold®	++	++	+++++	++	+++
MEM-a + FBS (qualified for MSC)	+	+	+++++	+	+

+++++ excellent performance
+ poor performance

MSC NutriStem® XF Medium

A Serum-Free, Xeno-Free Culture System

Product Name	Cat. #	Storage
MSC NutriStem® XF Basal Medium	05-200-1	2-8 °C
MSC NutriStem® XF Supplement Mix	05-201-1	-20 °C
MSC NutriStem® XF PRF Phenol Red-Free Basal Medium	05-202-1	2-8 °C
NutriCoat™ Attachment Solution	05-760-1-15	15 to 25 °C
MSC Attachment Solution	05-752-1	2-8 °C

Xeno-free, serum-free culture system, specially designed to support the growth of hMSCs from various sources.

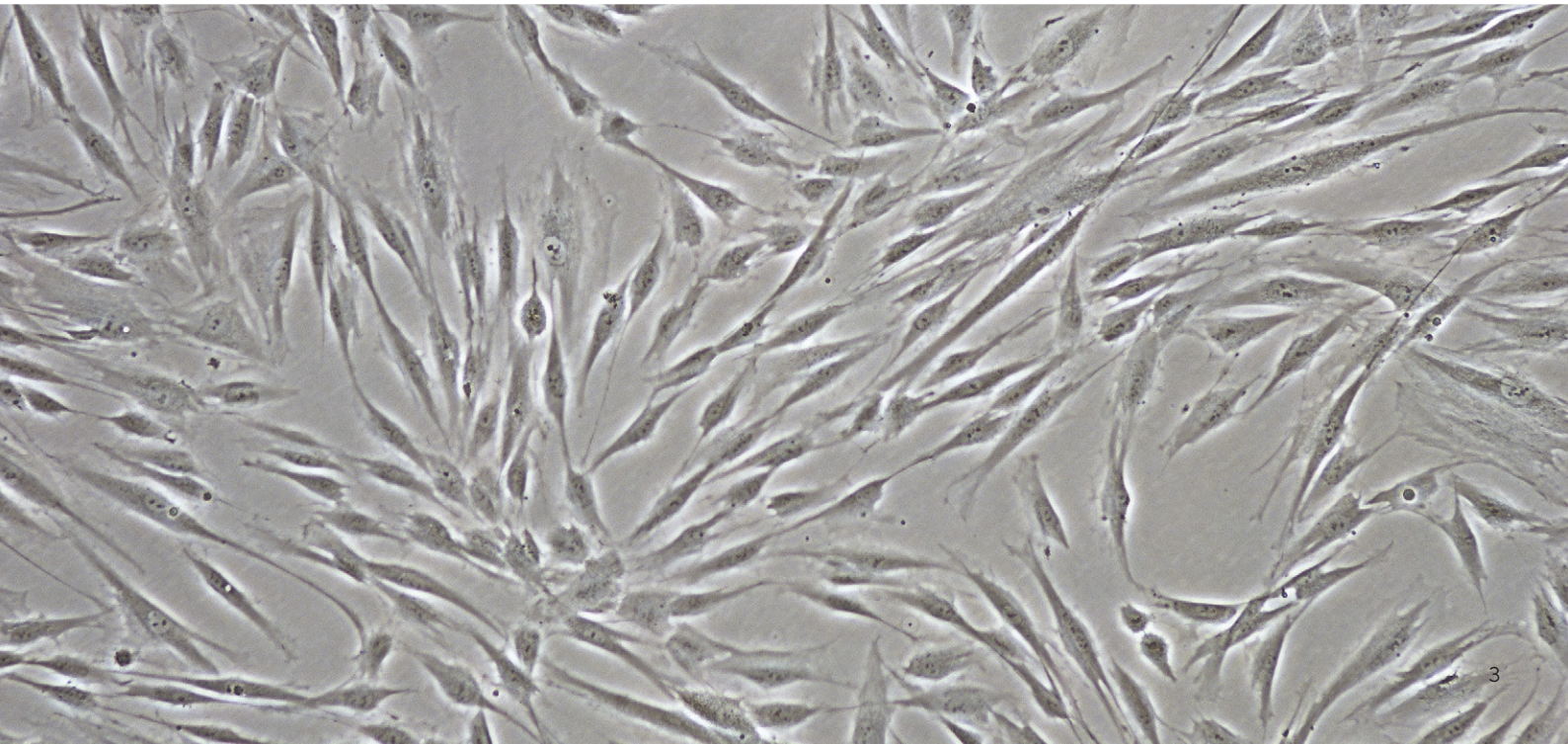
Advantages

Excellent performance
- Superior isolation and cell growth
- Superior maintenance of hMSC characteristics

Suitable for research and clinical applications
- Produced under cGMP conditions
- FDA drug master file available
- Used in clinical trials worldwide

Defined, serum-free, xeno-free medium
- Reproducible and consistent results throughout experiments
- Batch-to-batch consistency
- Save time and money: no need to prequalify FBS lots

Flexible medium
- Customization available
- Suitable for various MSC sources (i.e bone marrow, adipose tissue, cord tissue, placenta, dental pulp)

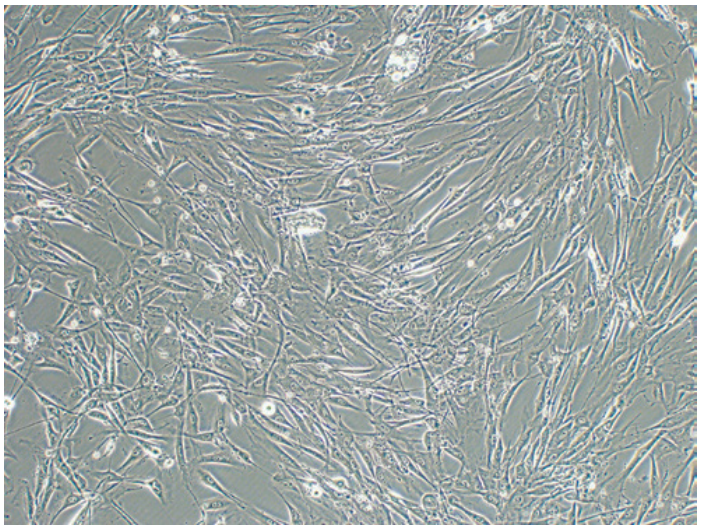


Isolation

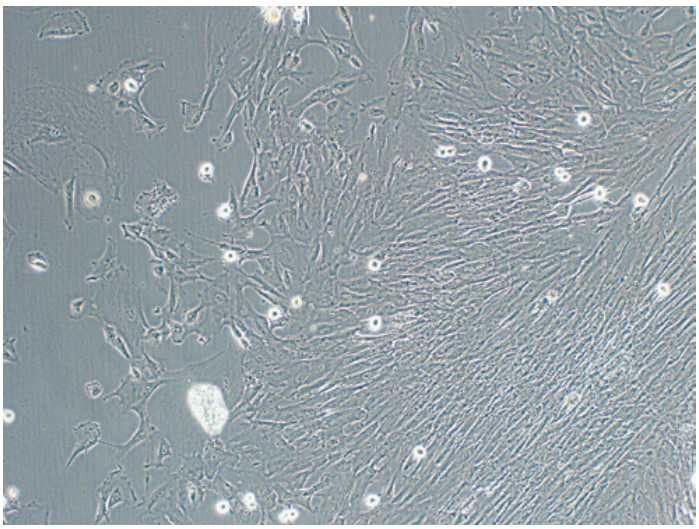
hMSCs from various sources (PL-MSCs, AT-MSCs, WJ-MSCs, BM-MSCs) can be efficiently isolated using MSC NutriStem® XF medium on pre-coated dishes. Addition of 2-2.5% human AB serum may be required for certain tissues. Using MSC NutriStem® XF for isolation of hMSC enhances purity of MSC populations in earlier passages and increases the number of hMSC in comparison to FBS-containing medium.

PL-MSCs

Figure 1:
hMSCs were isolated from frozen crude placenta under SF, XF culture conditions (MSC NutriStem® XF medium on pre-coated plates with MSC Attachment Solution, without supplementation of human AB serum) and in medium containing FBS. Representative images (×40) taken 11 days post initial isolation (P0).

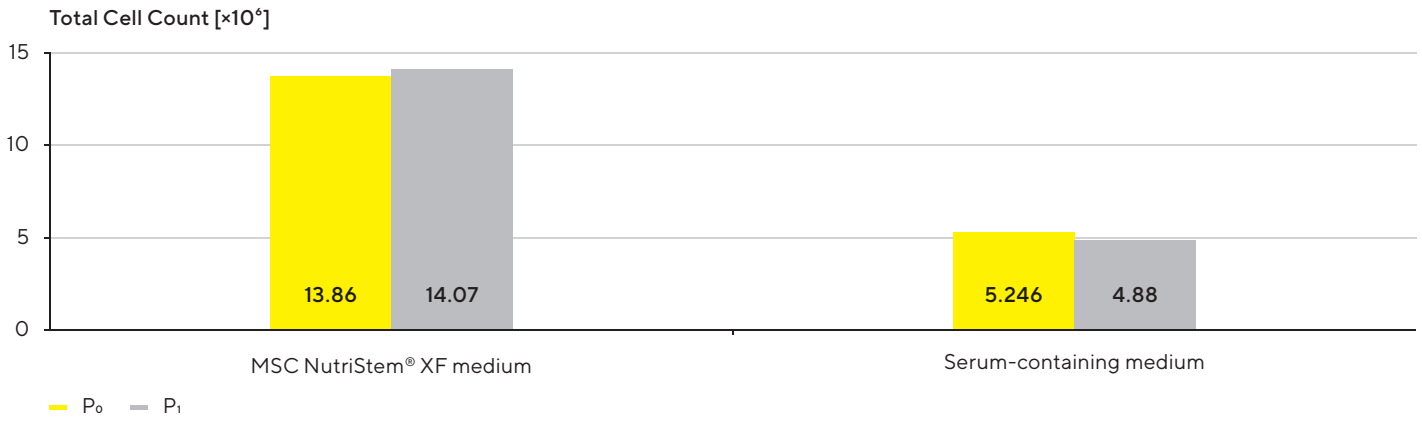


MSC NutriStem® XF medium



Serum-containing medium

Figure 2:
Comparison of PL-hMSC isolation from crude placenta 17 days post initial seeding (P0) in each medium. Quantity of viable cells, measured by trypan blue exclusion assay.

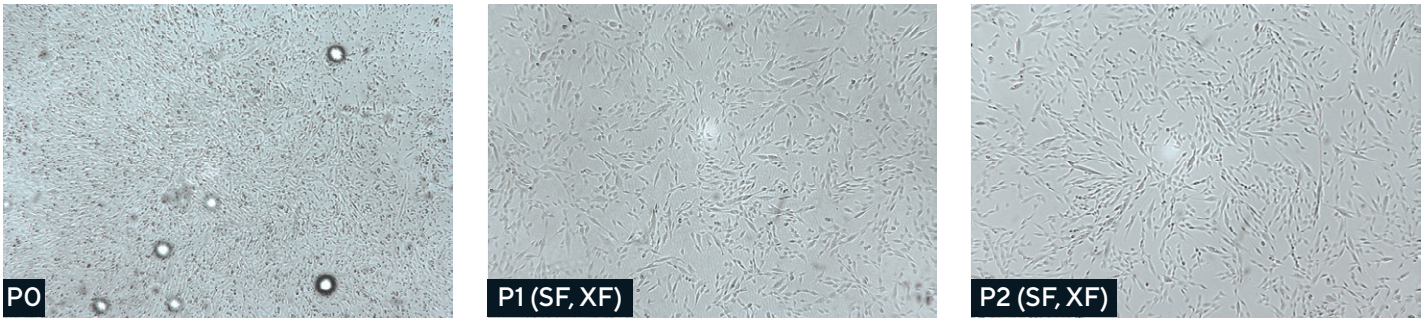


AT-MSCs

Figure 3:
Human AT-MSCs were seeded in MSC NutriStem® XF medium and supplemented with 2% human AB serum on pre-coated plates with MSC Attachment Solution for the initial isolation and expansion (P0).
The cells were cultured to 70-80% confluence before being sub-cultured. Further passages (P1-2) were done under SF, XF culture conditions, utilizing MSC NutriStem® XF culture medium on pre-coated dish.

A. Representative images taken 4 days post initial seeding (P0) and 3 days post P1 and P2.
B. Immunophenotyping results of AT-MSCs at passage 2 using FACS analysis.

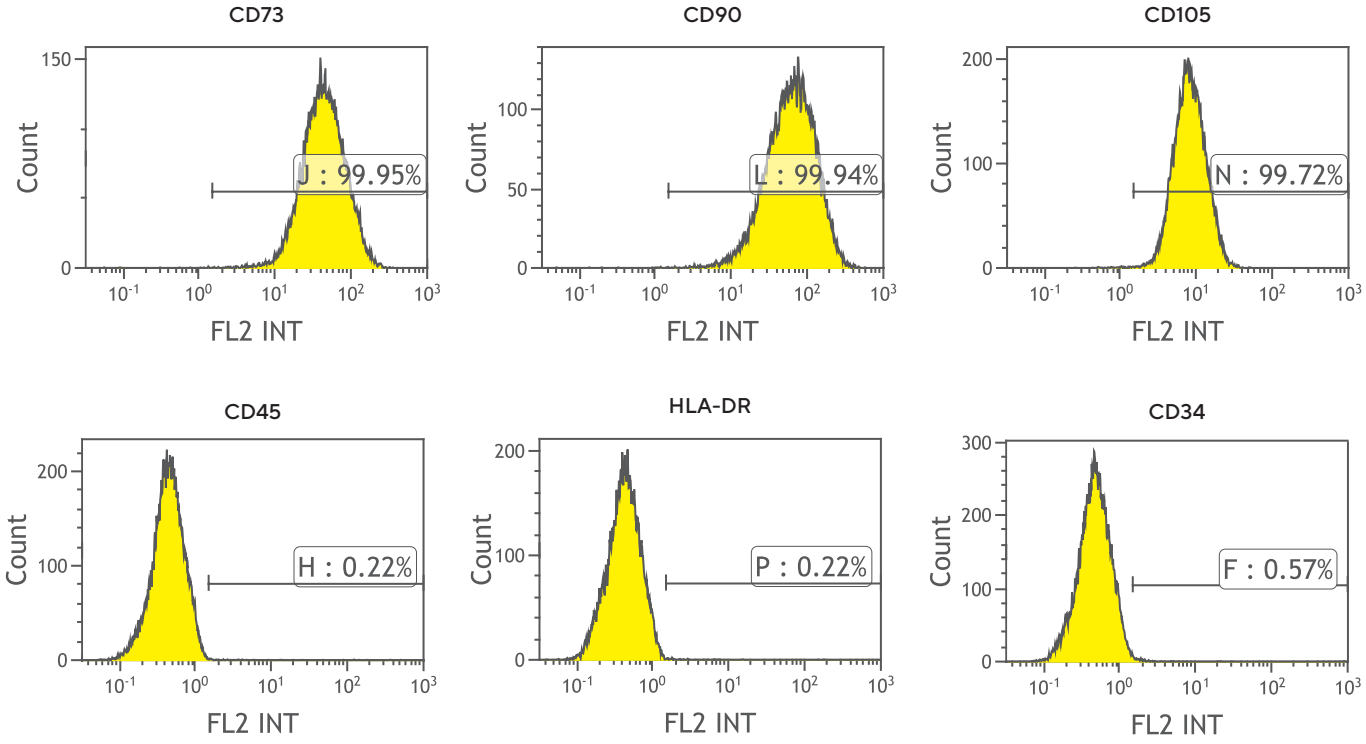
A



with human AB serum

without serum

B

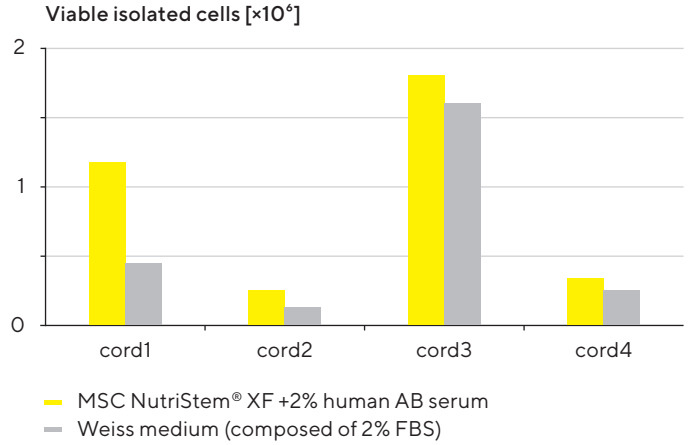


WJ-MSCs

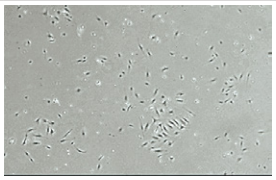
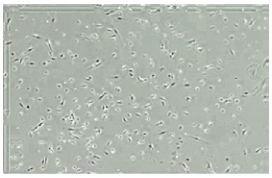
Figure 4:
WJ-MSCs were initially isolated from 4 independent human umbilical cords utilizing MSC NutriStem® XF medium supplemented with 2% human AB serum on pre-coated plates with MSC Attachment Solution in comparison to serum-containing medium.

A. Comparing the amount of viable cells – passage 0. Cell count was measured by trypan blue exclusion assay.
B. Representative images (×40) of cord 4 taken on day 2 post initial isolation in each medium, and cell count results of day 7 post initial isolation.

A



B

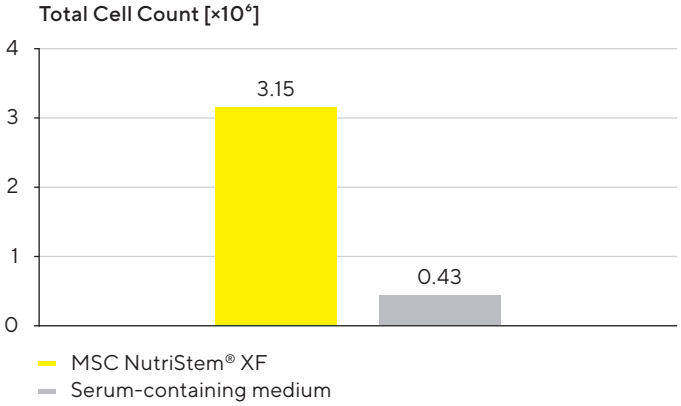
	Day 2 post initial isolation	Day 7 post initial isolation	
		Total cell count	
	Representative images	Live	Dead
MSC NutriStem® XF medium (supplemented with 2% human AB serum)		395,000	15,000
Weiss medium (composed of 2% FBS)		295,000	75,000

BM-MSCs

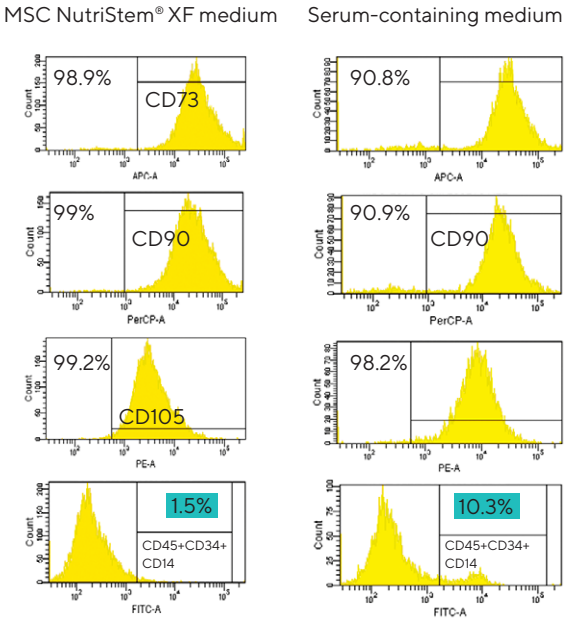
Figure 5:
Comparison of BM-MSC isolation from fresh BM utilizing MSC NutriStem® XF and serum-containing medium (11-day assay)

A. Cell count was measured by trypan blue exclusion assay.
B. Immunophenotype using FACS analysis.

A



B



Key References

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- Cai, Zhen, et al. Chondrogenesis of Human Adipose-Derived Stem Cells by In Vivo Co-graft with Auricular Chondrocytes from Microtia. Aesthetic plastic surgery 39.3 (2015): 431-439.
- S.H. Mei, et al. Isolation and large-scale expansion of bone marrowderived mesenchymal stem cells with serum-free media under GMP-compliance. Cytotherapy, Volume 16, Issue 4, Supplement, Page S111, April 2014
- Y.Lopez, M. Weiss, et al. Identification of Optimal Conditions for Generating MSCs for Preclinical Testing: Comparison of Three Commercial Serum-Free Media and Low-Serum Growth Medium. From 18th ISCT Annual Meeting, Seattle, USA, 2012.

Expansion

Superior Proliferation of hMSCs

hMSCs cultured in MSC NutriStem® XF medium exhibit higher proliferation rate and long term growth in comparison to competitors' media.

BM-MSCs

Figure 6:
Human BM-MSCs were cultured in MSC NutriStem® XF medium in comparison to commercial SF and serum-containing media. Initial seeding was 5000 cells/cm² for each of the tested media (day 0). Cells were counted daily by trypan blue exclusion assay.

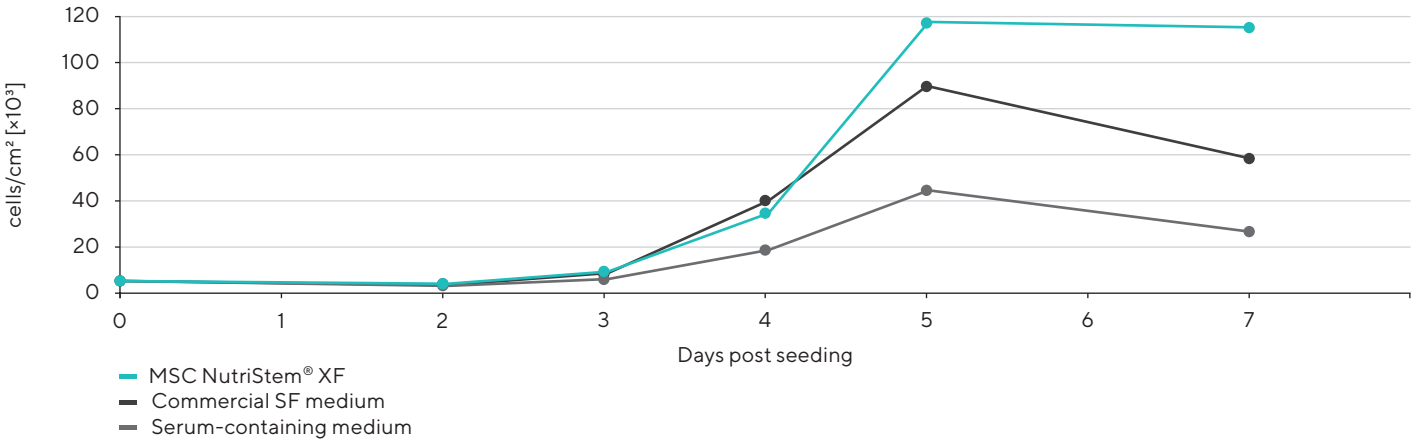
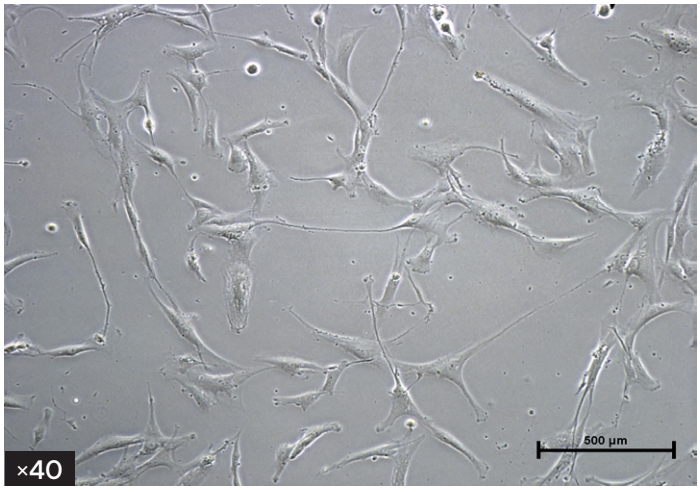
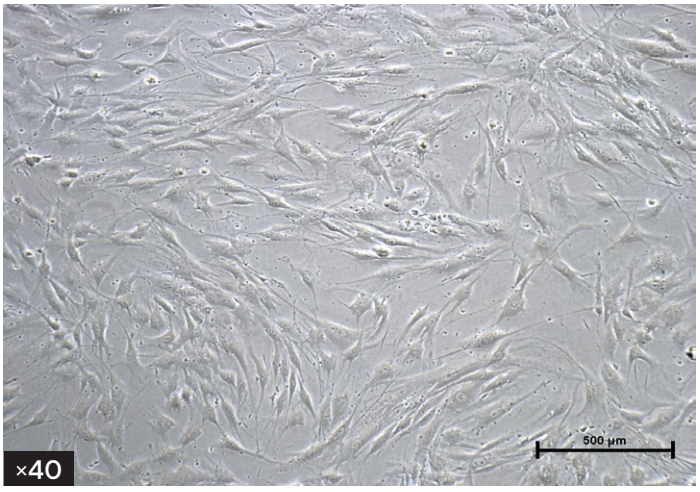


Figure 7:
Expansion of BM-MSCs in MSC NutriStem® XF medium and FBS-containing medium.
Initial seeding was 5000 cells/cm² for each of the tested media (day 0). Images were taken at day 3 post seeding.



Serum-containing medium



MSC NutriStem® XF medium

AT-MSCs

Figure 8:
Expansion of human AT-MSCs in MSC NutriStem® XF medium and commercially available XF, SF, and serum-containing media. Initial seeding was 5000 cells/cm² for each of the tested media (day 0). Cells were counted at day 3 in each passage.

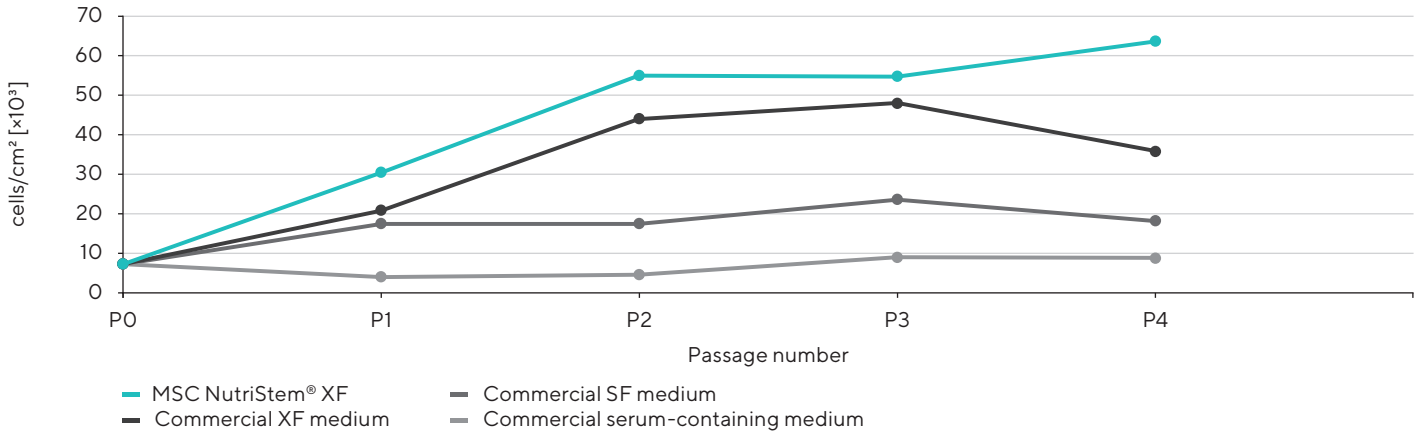
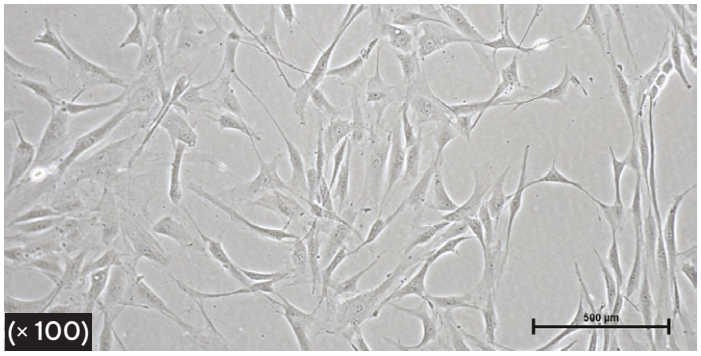
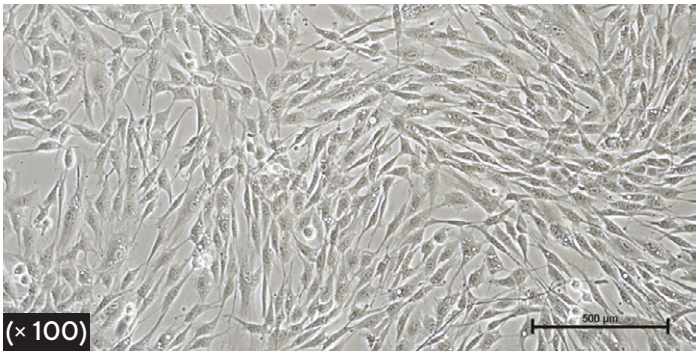


Figure 9:
Expansion of AT-MSCs in MSC NutriStem® XF medium in comparison to serum-containing medium.
Initial seeding was 6000 cells/cm² for each of the tested media (day 0). Images were taken 3 days post initial culture.



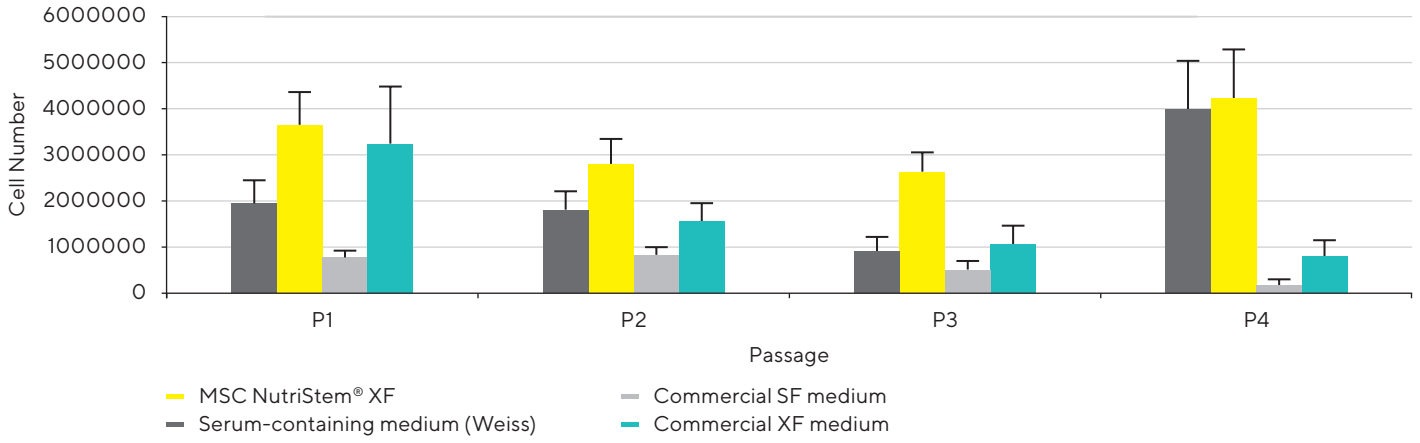
Serum-containing medium



MSC NutriStem® XF medium

WJ-MSCs

Figure 10:
Human WJ-MSCs from 9 different donors expanded for 4 passages in MSC NutriStem® XF medium in comparison to serum-containing medium and commercial SF and XF media. Cell proliferation was assessed by cell count using a trypan blue exclusion assay.

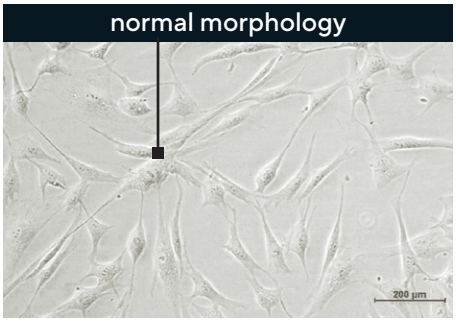


Cell Morphology

Typical fibroblast-like cells morphology was obtained when using MSC NutriStem® XF medium.

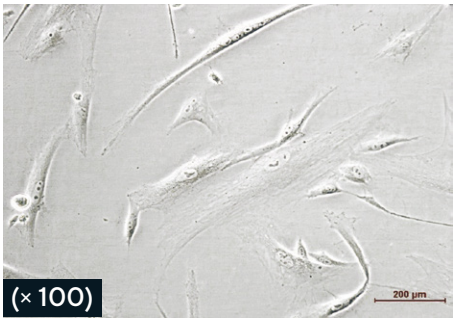
AT-MSCs

Figure 11:
Expansion of human AT-MSCs in MSC NutriStem® XF medium, competitor XF medium and competitor SF medium (day 0). Initial seeding was 5000 cells/cm² for each of the tested media. Images (×200) were taken 3 days post equal seeding (2 passages in each medium).



MSC NutriStem® XF
15×10⁴ cells/well

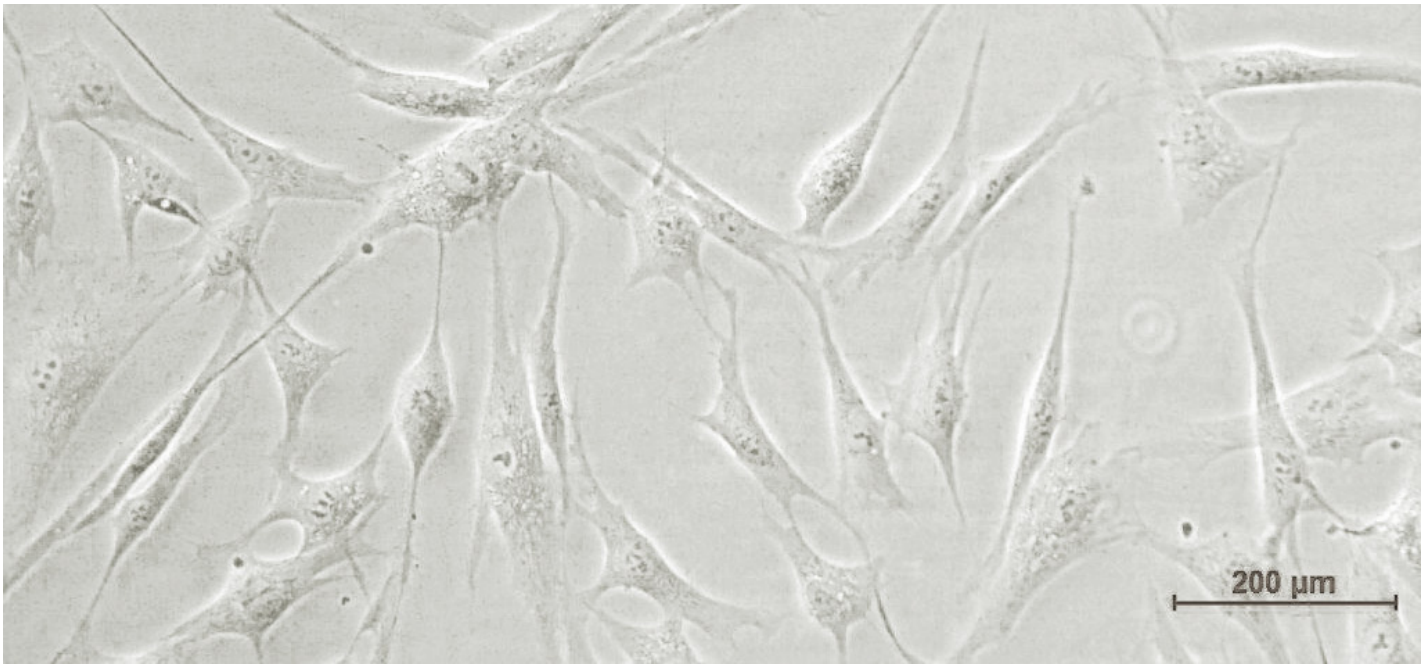
Commercial XF medium
11×10⁴ cells/well



Commercial SF medium
4×10⁴ cells/well

BM-MSCs

Figure 12:
Expansion of human BM-MSCs in MSC NutriStem® XF medium. Initial seeding was 5000 cells/cm² (day 0). Image was taken 4 days post passage 1 (×100). Typical "shoallike" pattern culture morphology is observed.



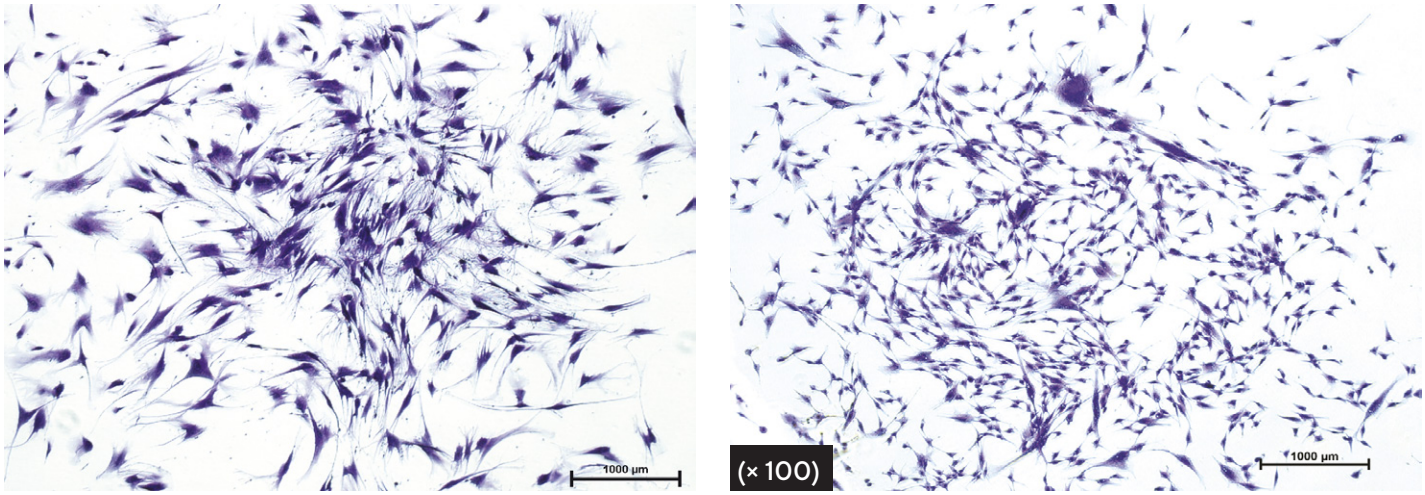
Self-Renewal Potential

hMSCs cultured in MSC NutriStem® XF medium maintain their selfrenewal potential.

BM-MSCs

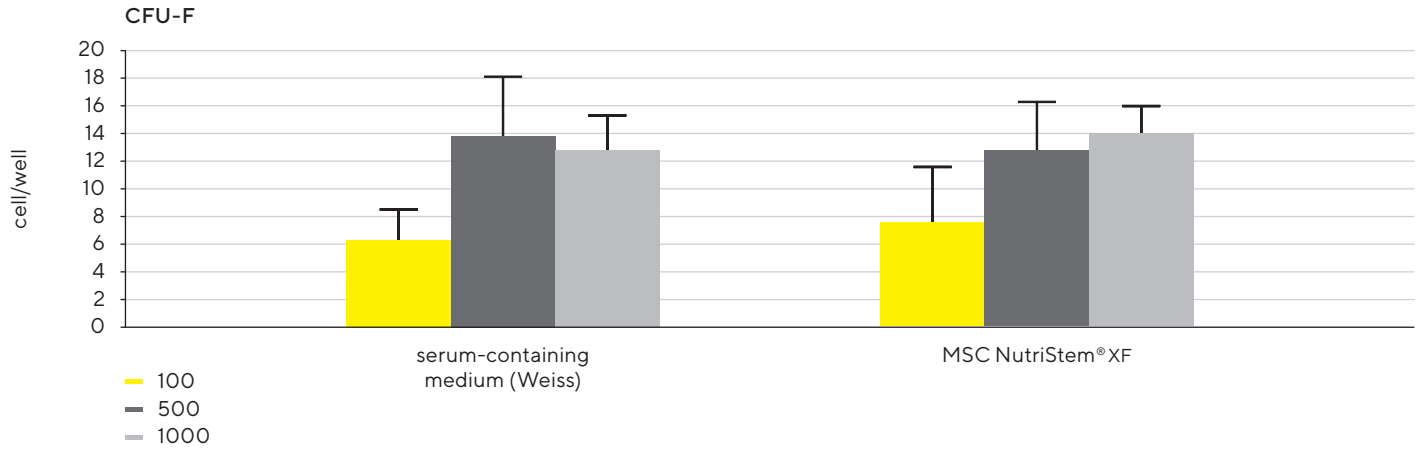
AT-MSCs

Figure 13:
Human AT and BM MSCs expanded in MSC NutriStem® XF medium for 3-5 passages prior to 14 day CFU-F assay. Representative images of colonies stained with 0.5% crystal violet (×100).



WJ-MSCs

Figure 14:
CFU-F assay of human WJ-MSCs expanded for 5 passages in MSC NutriStem® XF medium and Weiss medium (2% FBS) in 3 different seeding concentrations.



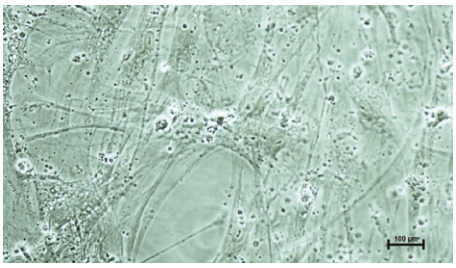
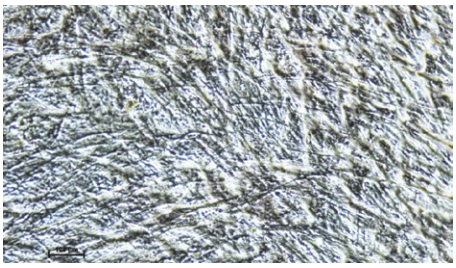
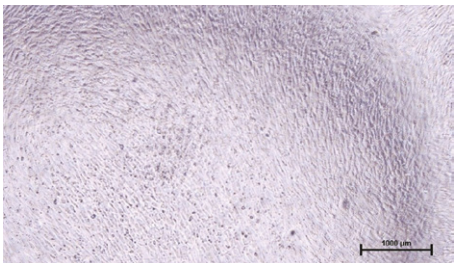
Differentiation Potential

hMSCs cultured in MSC NutriStem® XF medium maintain their trilineage differentiation potential.

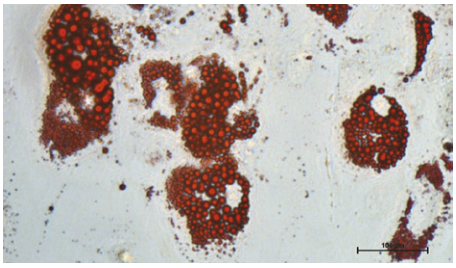
Figure 15:
Human BM and AT MSCs were expanded in MSC NutriStem® XF medium for 3-5 passages prior to differentiation. Representative images of stained Adipocytes (Oil Red O), Osteocytes (Alizarin red) and Chondrocytes (Alician blue). The control images show cells which were cultured in MSC NutriStem® XF medium for the whole term. Staining was not obtained in the control cells.

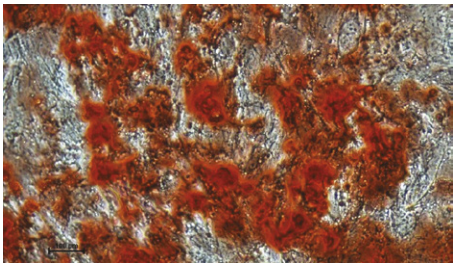
BM-MSCs

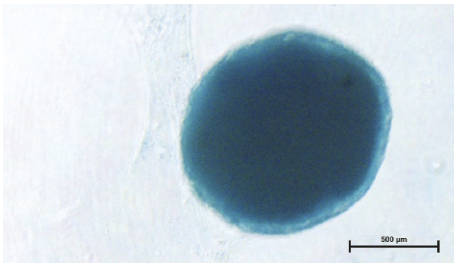
Control

Differentiation

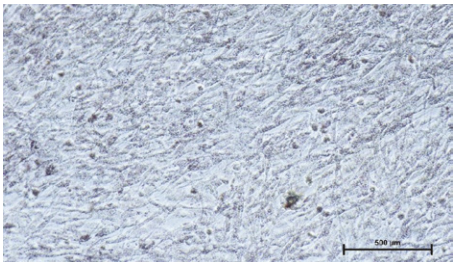
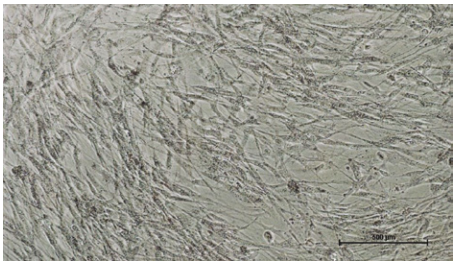
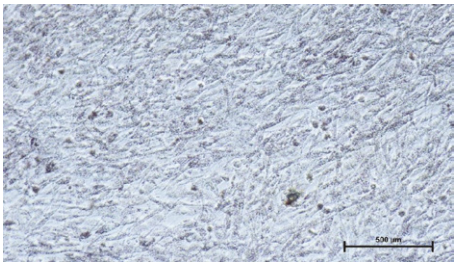

Adipocytes - Oil red O


Osteocytes - Alizarin red

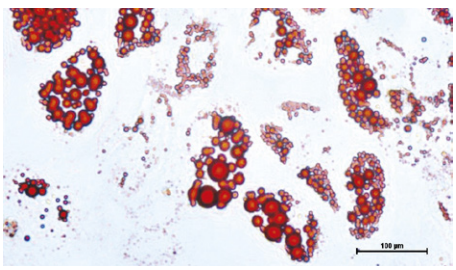

Chondrocyte - Alcian blue

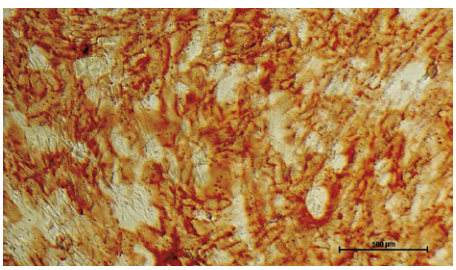
AT-MSCs

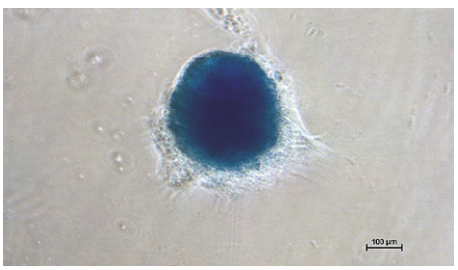
Control

Differentiation


Adipocytes - Oil red O


Osteocytes - Alizarin red


Chondrocyte - Alcian blue

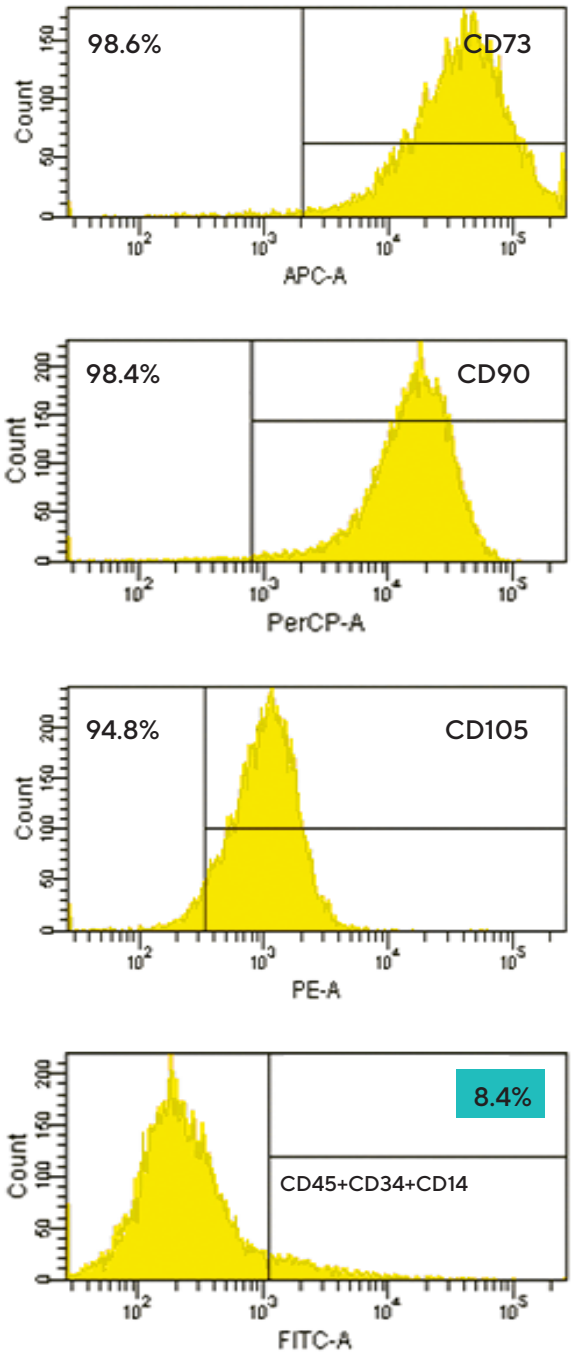
Surface Markers Profile

hMSCs expanded in MSC NutriStem® XF medium kept their classical profile of MSC markers; stained for MSC positive surface markers and did not stain for hematopoietic markers.

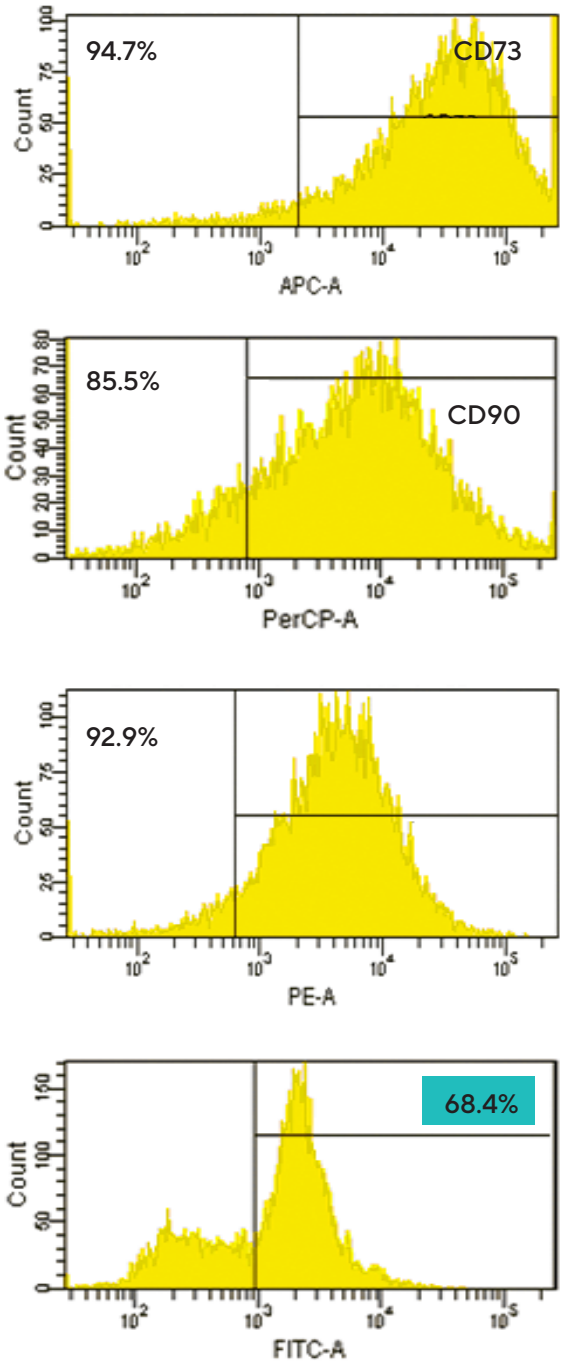
PL-MSCs

Figure 16:
Immunophenotype results of PL-MSCs after culturing in each medium. **Purer hMSC population is achieved using MSC NutriStem® XF medium.**

MSC NutriStem® XF



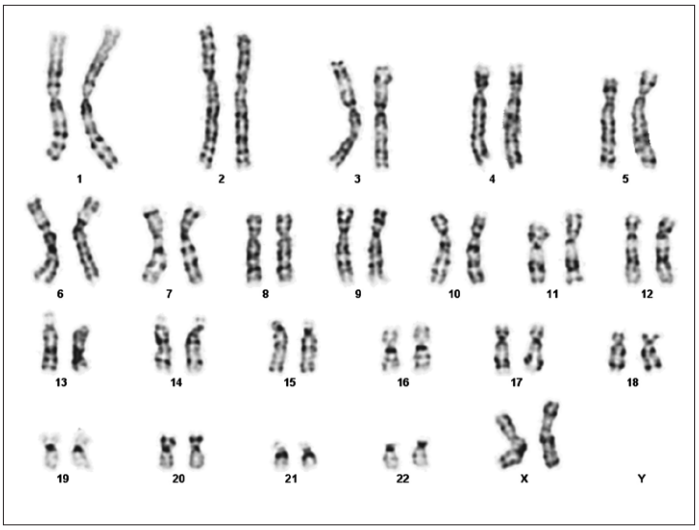
Serum-containing medium



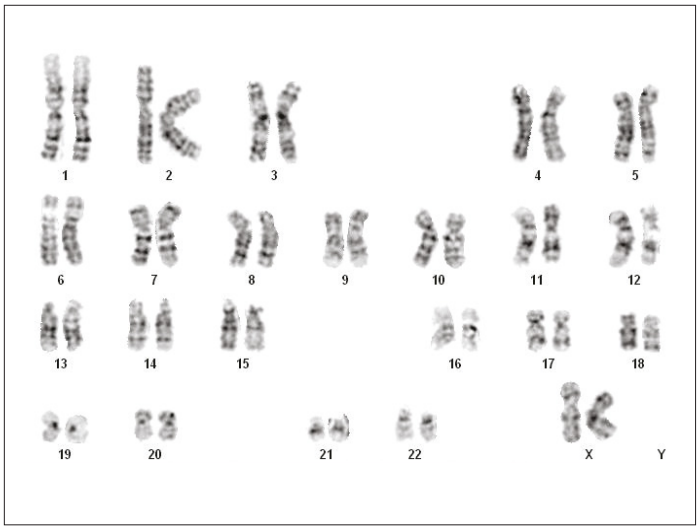
Karyotyping

Normal karyotypes of BM-MSCs (46, XY) AT-MSCs (46,XX) and CT-MSCs (46, XX) were observed after long term culturing in MSC NutriStem® XF medium.

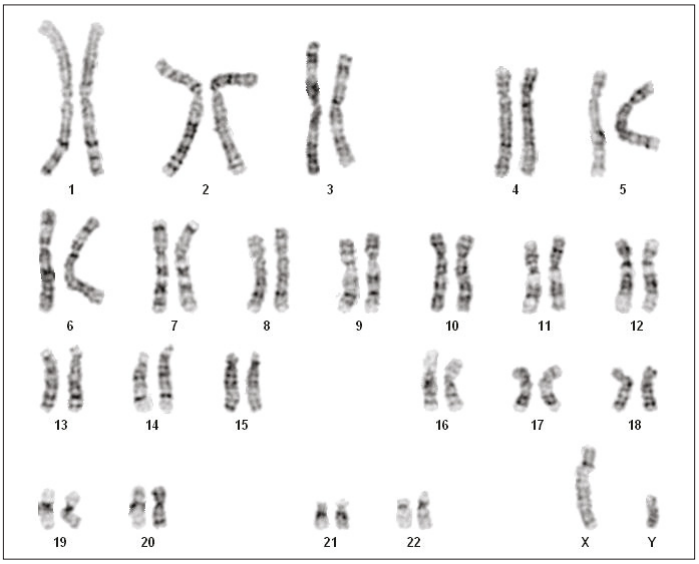
Figure 17:
G-banding karyotyping analysis of hMSCs from various sources expanded for 4-9 passages in MSC NutriStem® XF medium. hMSCs cultured in MSC NutriStem® XF medium maintain genomic stability.



AT-MSCs
P4 PD15
46, XX



CT-MSCs
P6 PD18
46, XX

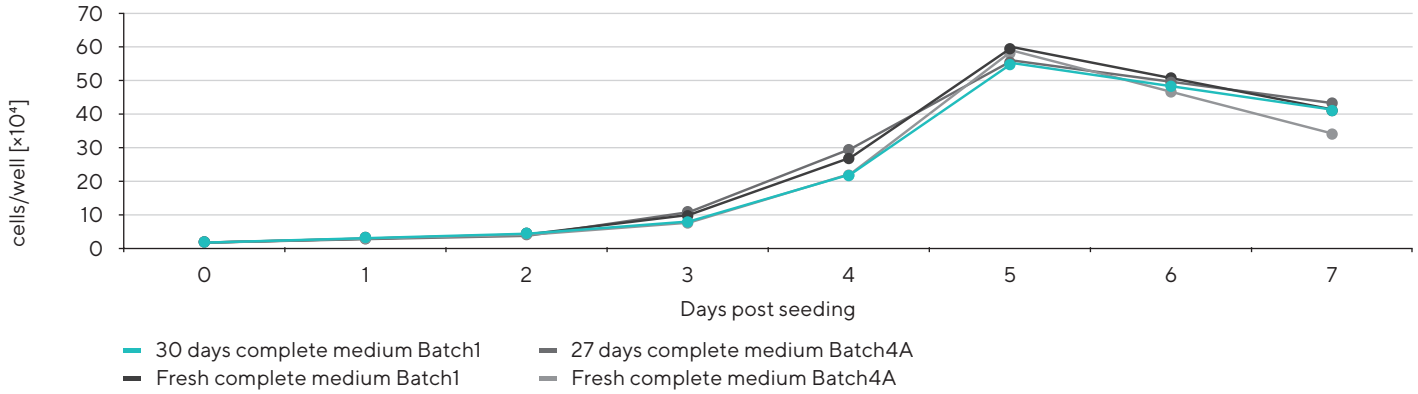


BM-MSCs
P9 PD20
46, XY

MSC NutriStem® XF Complete Medium Stability

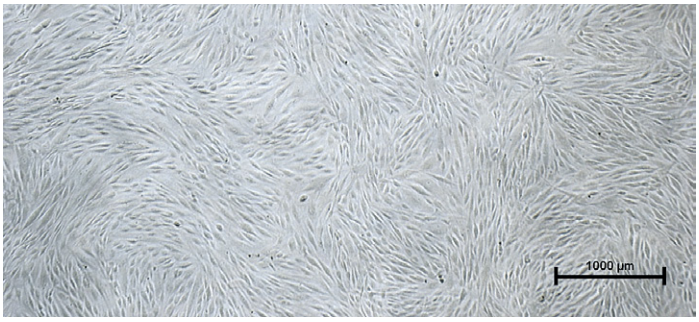
The complete MSC NutriStem® XF medium is stable for 30 days at 2-8 °C.
No significant differences of hMSC proliferation were observed between fresh and 30 days old complete medium.

Figure 18:
Growth curve of BM-MSCs cultured in MSC NutriStem® XF medium. Results were taken from 2 batches: batch No. 1: 7.5 months from production and batch 4A: 2.5 months from production. A complete medium was prepared 30 & 27 days before seeding (stored at 2-8 °C) or freshly prepared. Initial seeding was 5000 cells/cm² for each of the tested media (day 0). Cells were counted daily by trypan blue exclusion assay.

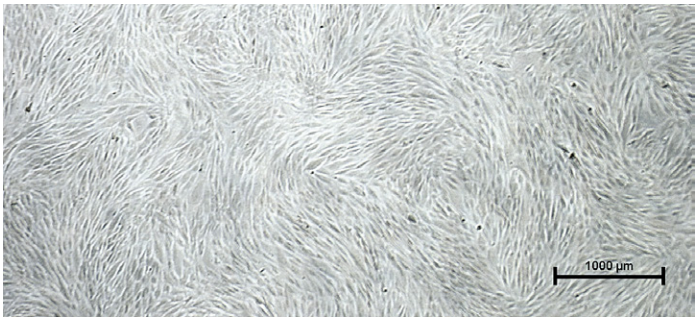


MSC NutriStem® XF complete medium

Figure 19:
A complete medium was prepared freshly, or 30 days before seeding (stored at 2-8°C). Initial seeding was 5000 cells/cm² for each of the tested media (day 0). Images were taken 4 days after equal split 1 (P2).



Fresh (×40)
40×10³ cells/cm²



30 Days (×40)
42×10³ cells/cm²

Key References

Clinical Trials
1. McIntyre, L.A, et. al. Cellular Immunotherapy for Septic Shock. A Phase I Clinical Trial. Ameri. J. of Res. and Critical Care Medicine, Vol. 197, No. 3, 2018.
2. Schlosser, K., et al. Effects of MSC Treatment on Systemic Cytokine Levels in a Phase 1 Dose Escalation Safety Trial of Septic Shock Patients. Critical Care Medicine. DOI 10.1097/CCM.0000000000003657

Publications
1. Valitsky, M., et al.2019. Int. J. Mol. Sci. 20(7), 1793; https://doi. org/10.3390/ijms20071793.
2. Narbute, K., et al. 2019. STEM CELLS TRANSLATIONAL MEDICINE, pp. 1-10.
3. Jonavice, U., et al. 2019. J. of Tissue Engine. and Regen. Med.
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11. Khan, S., et al. 2018. Cytotherapy. 20. 5: S42. 10. Volz, A.C., et. al. 2018. 20. 4:576–588.
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16. Berger, L., et al. 2015. STEM CELLS. 34. 4:31 DEC.
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18. Zhen, C. et al. 2015. Aesth. 39.3: 431-439.
19. Pokrywczynska, M., et al. 2015. Archi. Immun. Therap. Exper.
20. Mei, S.H., et al. 2014. Cytotherapy. 16. 4:S111.
21. McVey, M., et al. 2016 Ameri. J. of Phys. ajplung-00369.
22. Lopez, Y., et al. 2012. ISCT Annual Meeting, Seattle, USA.
23. Foster, R. D., et al. 2011. Tissue Eng Meth. Part C. 17:7.

The above reference guide only represents a sample of citations for these products.

Coating-Free Options

MSC NutriStem® XF with CellBIND®

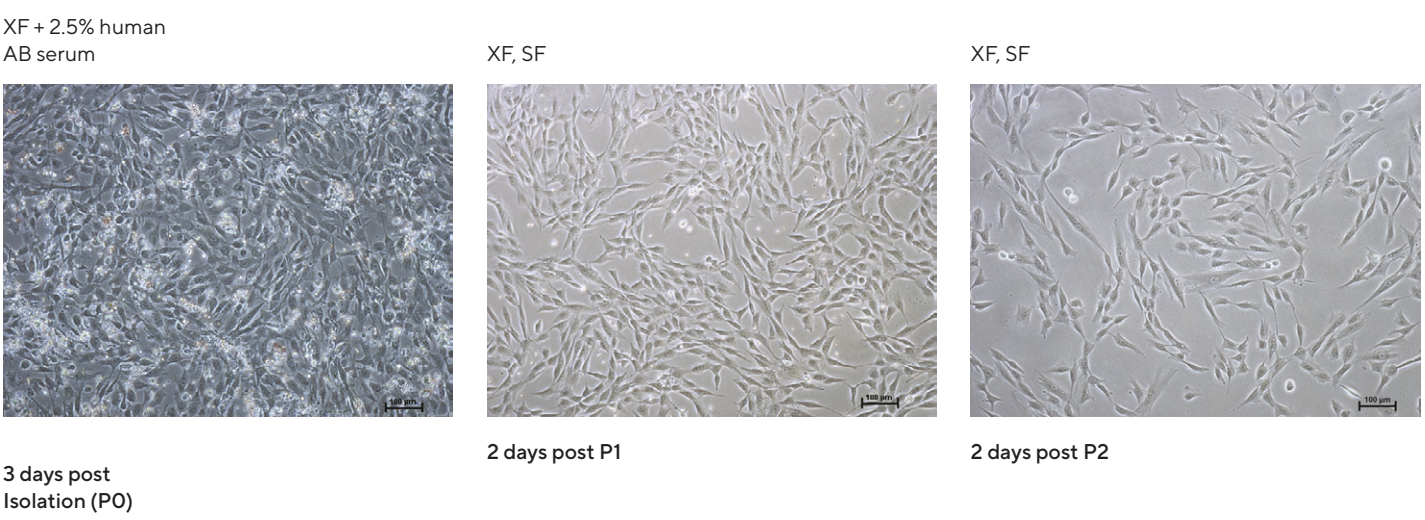
MSC MutriStem® XF medium shows superior performance in comparison to competitors media, using Corning CellBIND® surface with various tissues (no need for plate coating).

Isolation

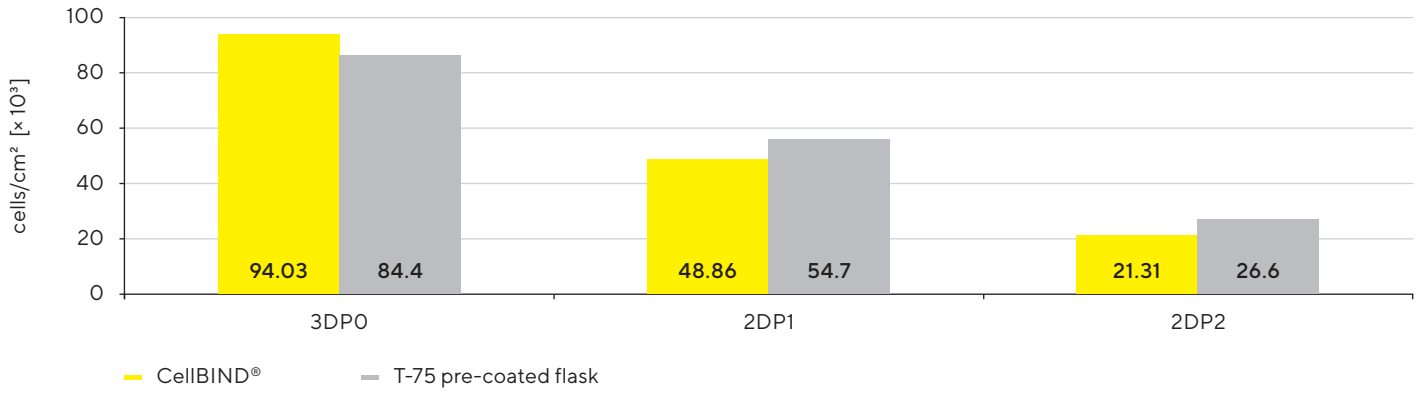
Figure 20:
Human AT-MSC isolation results using MSC NutriStem® XF medium and CellBIND®. 2.5% human AB serum was added only at P0.
A. Representative images (×100)
B. Quantity of viable cells.

Note: in comparison to the use of MSC NutriStem® XF medium with pre-coating procedure, similar proliferation rate is observed after seeding, however further passages using CellBIND® may lead to slightly reduced proliferation rate.

A



B



* CellBIND® is a registered trade mark of Corning.

Expansion

Morphology and Proliferation

Superior morphology and higher proliferation of hMSCs from various sources using MSC NutriStem® XF medium with CellBIND®.

Figure 21:
hMSCs were cultured in MSC NutriStem® XF medium and in commercial xeno-free media using CellBIND®. Representative images (×100) 3 days post-split 1.

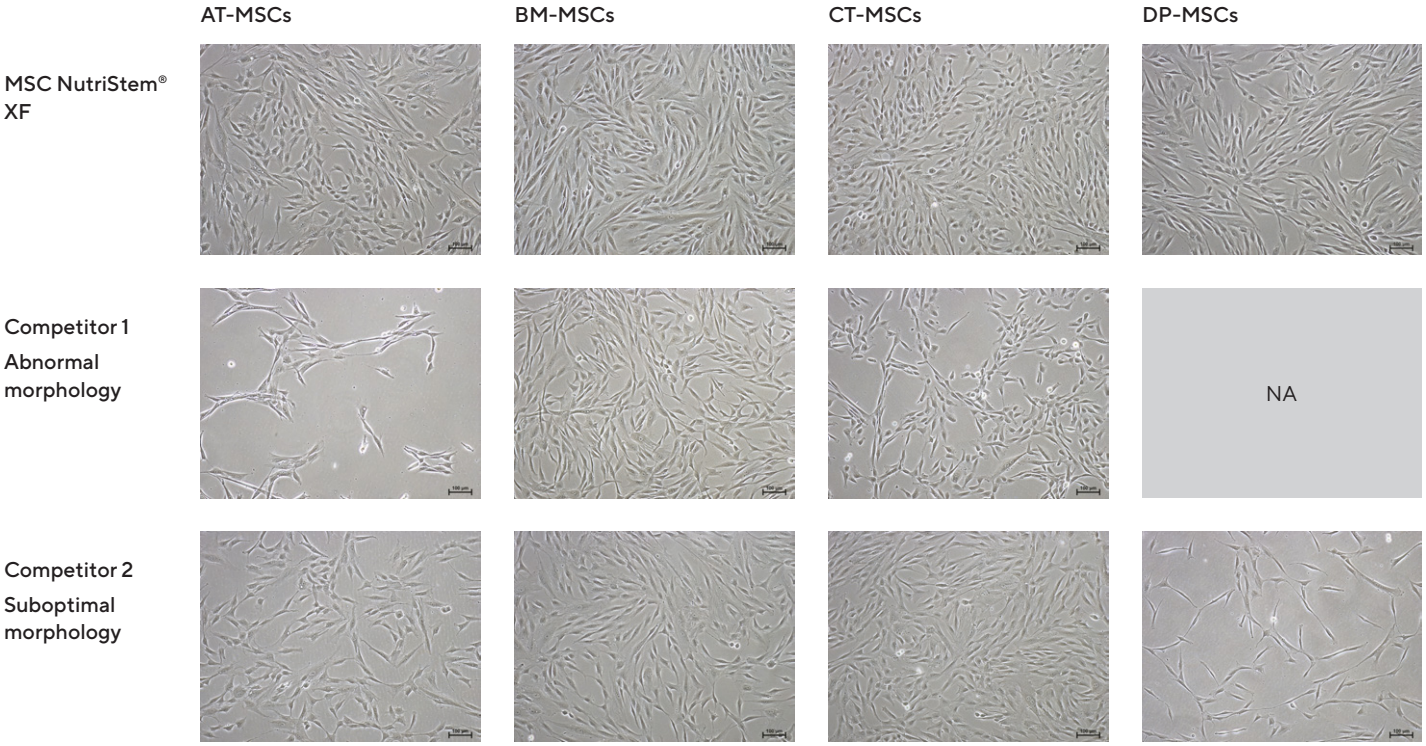
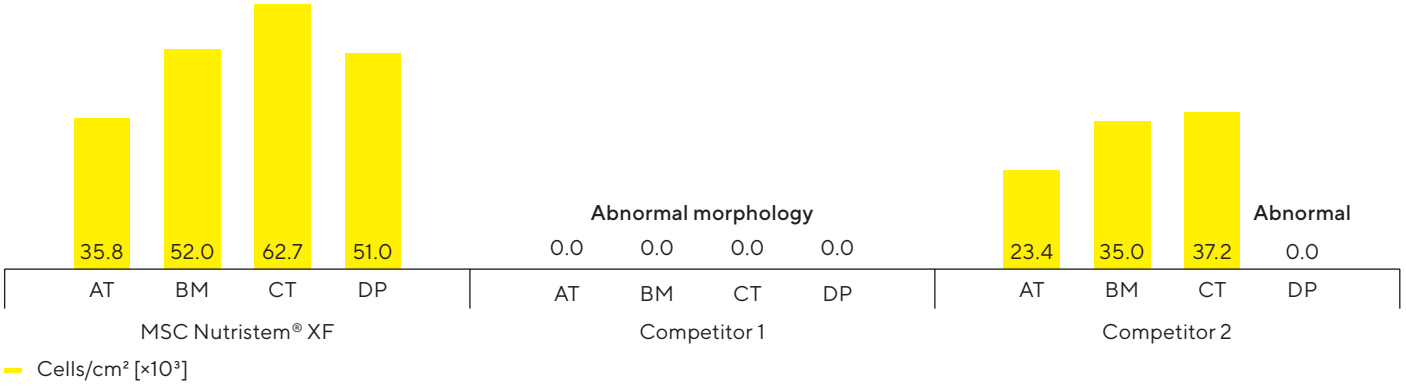


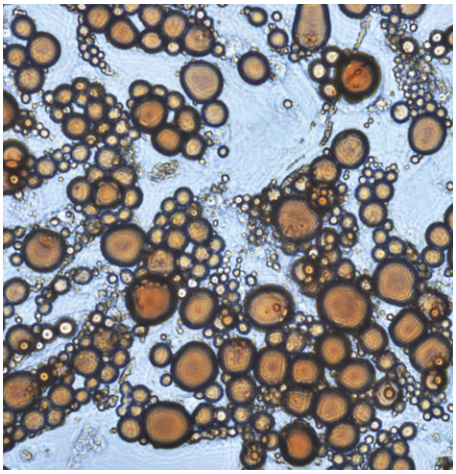
Figure 22:
hMSC proliferation in MSC NutriStem® XF and in commercial xeno-free media using CellBIND®.



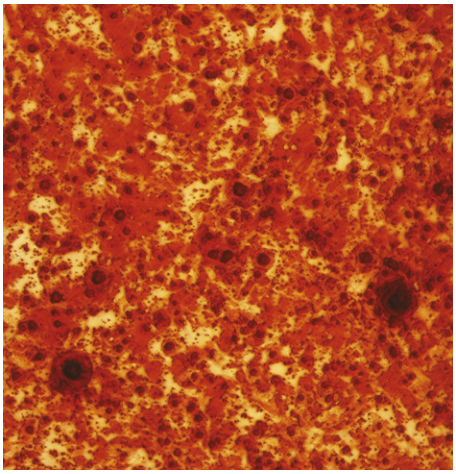
Differentiation Potential

hMSCs cultured in MSC NutriStem® XF medium using CellBIND® maintain their tri-lineage differentiation potential.

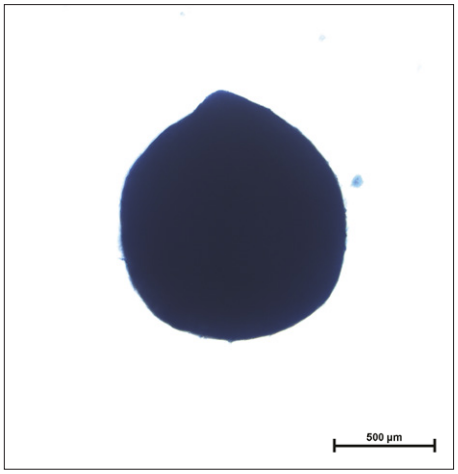
Figure 23:
Human AT-MSCs were isolated on CellBIND® uncoated plate in MSC NutriStem® XF medium +2% human AB serum followed by 2 passages in MSC NutriStem® XF medium w|o AB serum. The differentiation assay was done using MSCgo™ Adipogenic, MSCgo™ Osteogenic and MSCgo™ Chondrogenic media.



Adipocytes - Oil red O



Osteocytes - alizarin red

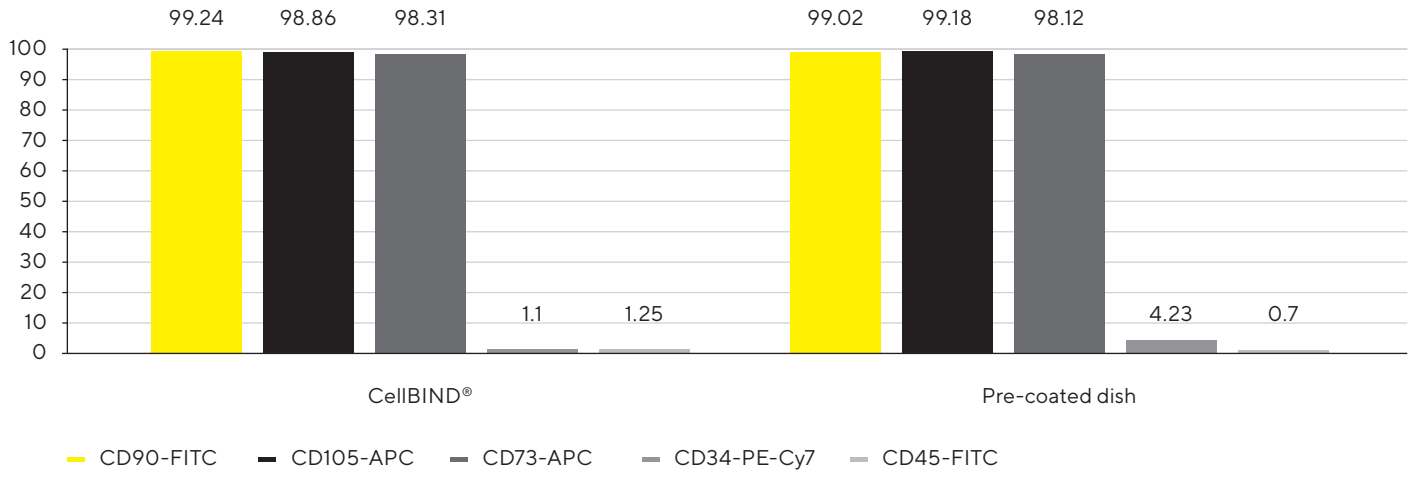


Chondrocytes - alcian blue

Marker Expression

hMSCs cultured in MSC NutriStem® XF medium using CellBIND® kept their classical profile of MSC markers.

Figure 24:
Flow cytometry analysis of human AT-MSCs after 2P post isolation in MSC NutriStem® XF medium, using CellBIND® and precoated dish. % expression -CD90+105+34 1:250; CD73+45 1:500.



MSC NutriStem® XF Medium With Human Platelet Lysate

MSC NutriStem® XF medium shows excellent performance in a xeno-free culture system with the addition of PLTGold® Human Platelet Lysate (no need for plate coating).

Morphology and Proliferation

Figure 25:
Adipose-derived MSCs expansion using Platelet Lysate. MSCs were expanded using a variety of supplements and the confluence was observed over time.

A. Cultures of MSCs using PLTMax® and PLTGold® showed the highest and fastest confluence;
B. Multiple batches of PLTGold® showed similar to identical growth rates

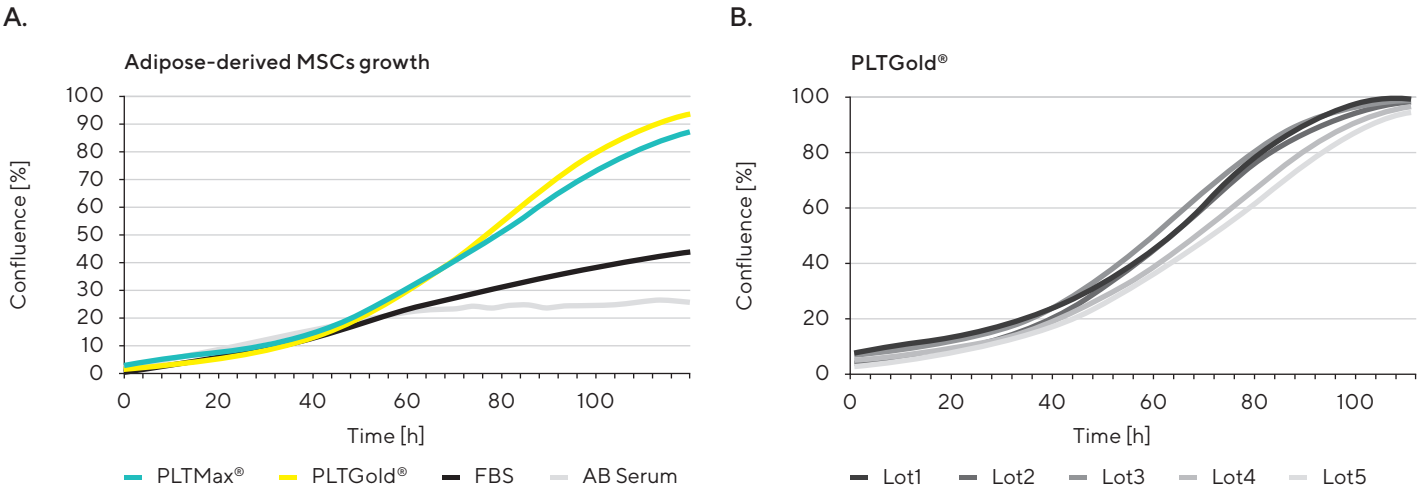


Figure 26:
Fold expansion of MSCs using MSC NutriStem® or A-MEM. MSCs grown using MSC NutriStem® and 5% PLTGold® showed 100-200 fold expansion higher than using Advanced-MEM over the course of 5 Passages.

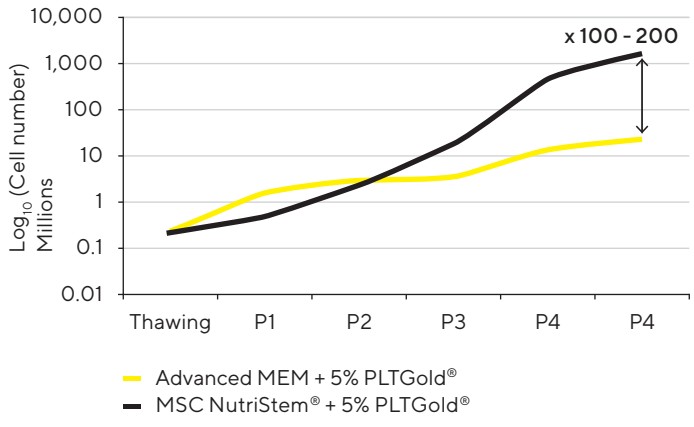
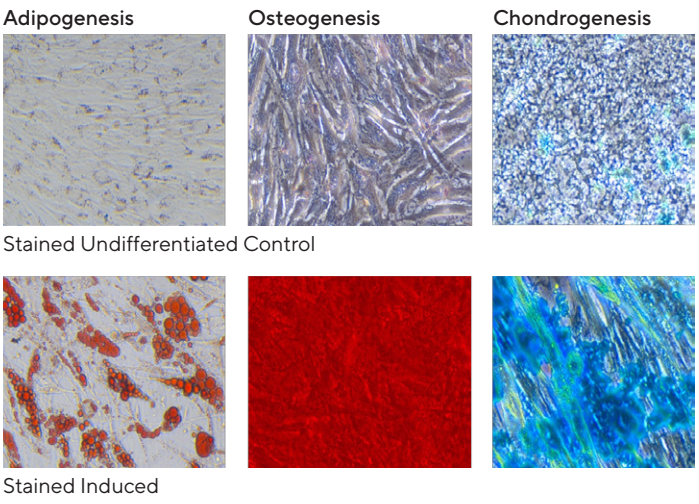


Figure 27:
Multipotency of MSCs grown with Platelet Lysate supplements. MSCs grown using Platelet Lysate show the ability to differentiate to adipocytes, osteocytes and chondrocytes.



Dissociation

Product Name	Cat. #	Storage
Recombinant Trypsin Solution without EDTA	03-078-1	15-30 °C
Recombinant Trypsin Solution with EDTA	03-079-1	15-30 °C

Recombinant Trypsin Solution is an ACF cell dissociation solution, designed as an alternative to porcine|bovine trypsin. The addition of EDTA usually accelerates the dissociation phase. The solutions do not contain any chymotrypsin, carboxypeptidase A, or other protease contaminant. Recombinant Trypsin Solution formulations were developed for efficient dissociation of adherent cell types from surfaces and tissues and were optimized for sensitive cells, such as hMSCs.

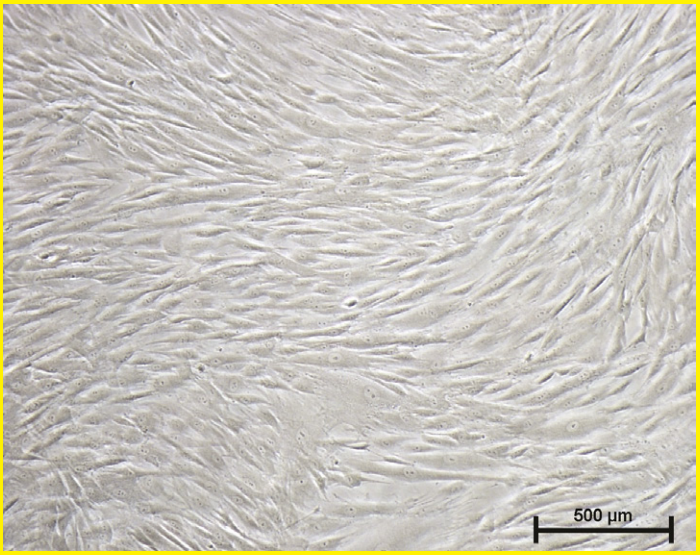
Advantages:

- Ready-to-use
- Non-animal or human origin
- Optimal for hMSCs (from a variety of sources), cultured in both SF and serum-containing systems
- Free from undesirable proteases such as carboxypeptidase A and chymotrypsin
- Eliminates contaminating activities found in bulk production of enzymes
- Storage: room temperature

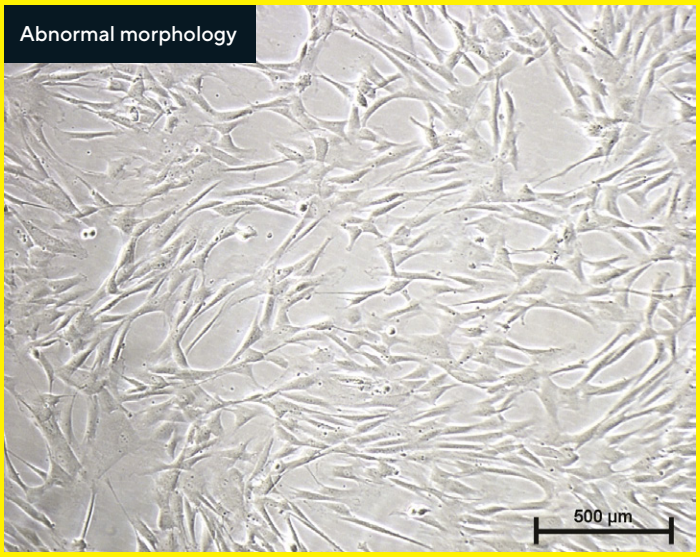
Neutralization of Recombinant Trypsin is achieved with MSC NutriStem® XF medium or Soybean Trypsin Inhibitor (SBTI) Cat. No.: 03-048-1.

The use of recombinant trypsin, rather than crude trypsin, is often essential for successful, long term growth of cells under SF culture conditions.

Figure 28:
Recovery of BM-MSCs after dissociation with both Recombinant Trypsin Solution and the common Trypsin EDTA Solution (porcine) following re-seeding in MSC NutriStem® XF medium on pre-coated plates. Representative images were taken on day 5 post-dissociation (×100).



Recombinant Trypsin Solution



Crude Trypsin EDTA Solution

Cryopreservation

Product Name	Cat. #	Storage
NutriFreez® D10 Cryopreservation Medium	05-713-1	2-8 °C
NutriFreez® D10 Cryopreservation Medium, w|o phenol red	05-714-1	2-8 °C

NutriFreez® D10 Cryopreservation Medium is a chemically defined, animal component-free and protein-free formulation for the cryopreservation of animal cells. The medium shows excellent performance, high cell viability and cell recovery after thawing and is suitable for hMSCs from various sources.

Advantages:

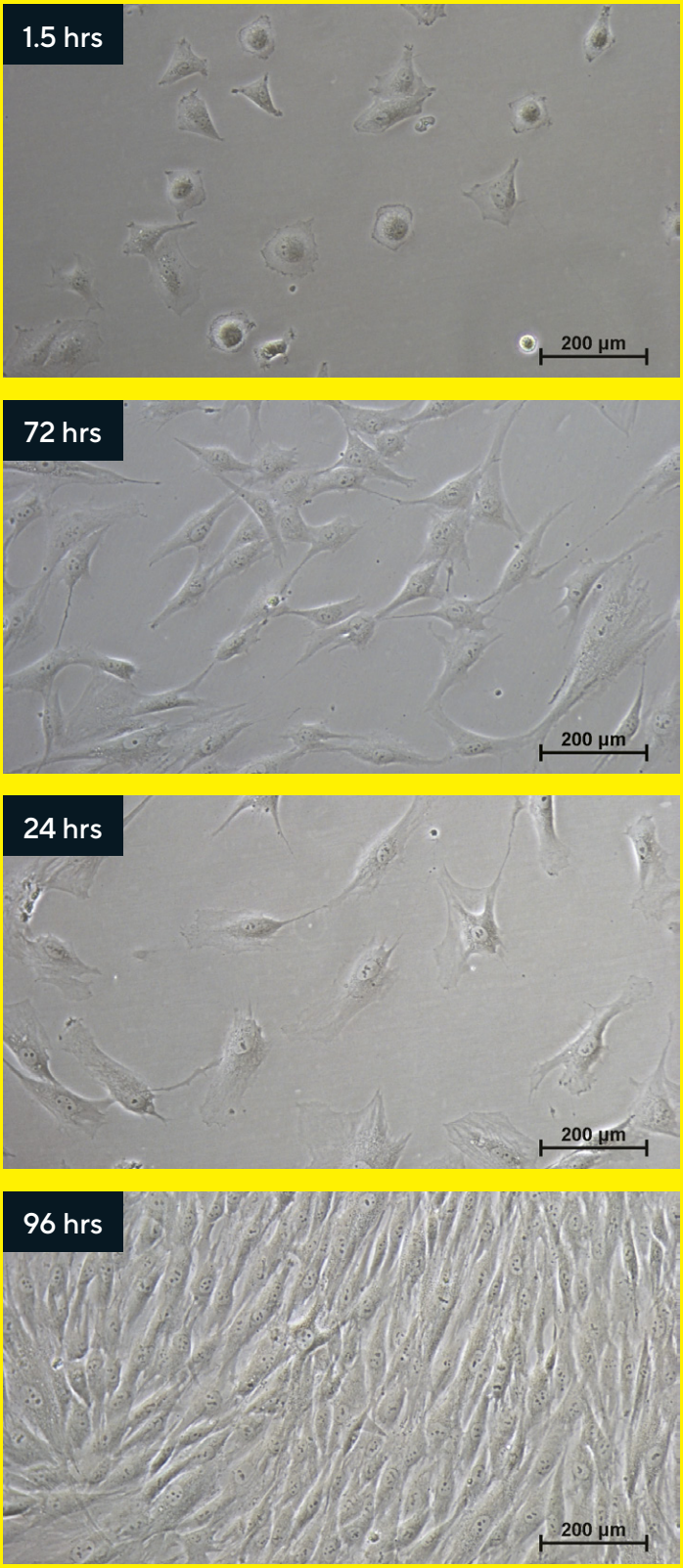
- A complete, ready-to-use solution (2-8 °C)
- Protein-free
- Animal components-free
- Suitable for hMSCs from various sources
- Suitable for cells cultured in both SF and serum-containing medium
- High cell viability and cell recovery after thawing

Cryopreservation of hMSCs using NutriFreez® D10 Cryopreservation Medium led to high viability and high recovery rate after thawing.

	Total cells [cells/ml]	Nonviable cells [cells/ml]	Viable cells [cells/ml]	Viability [%]
Test 1	9.36×10^5	3.97×10^4	8.96×10^5	**95.8**
Test 2	8.82×10^5	4.84×10^4	8.34×10^5	**94.5**

Human BM-MSCs (2 individual tests) were thawed and expanded in MSC NutriStem® XF medium, 15 months post cryopreservation.

Figure 29:
Recovery of human BM-MSCs after thawing procedure. Cells were frozen using NutriFreez® D10 Cryopreservation Medium, thawed and re-seeded in MSC NutriStem® XF medium on precoated plates. Representative images were taken at the indicated time points post-thawing (×200).



Differentiation

MSCgo™ Differentiation Media

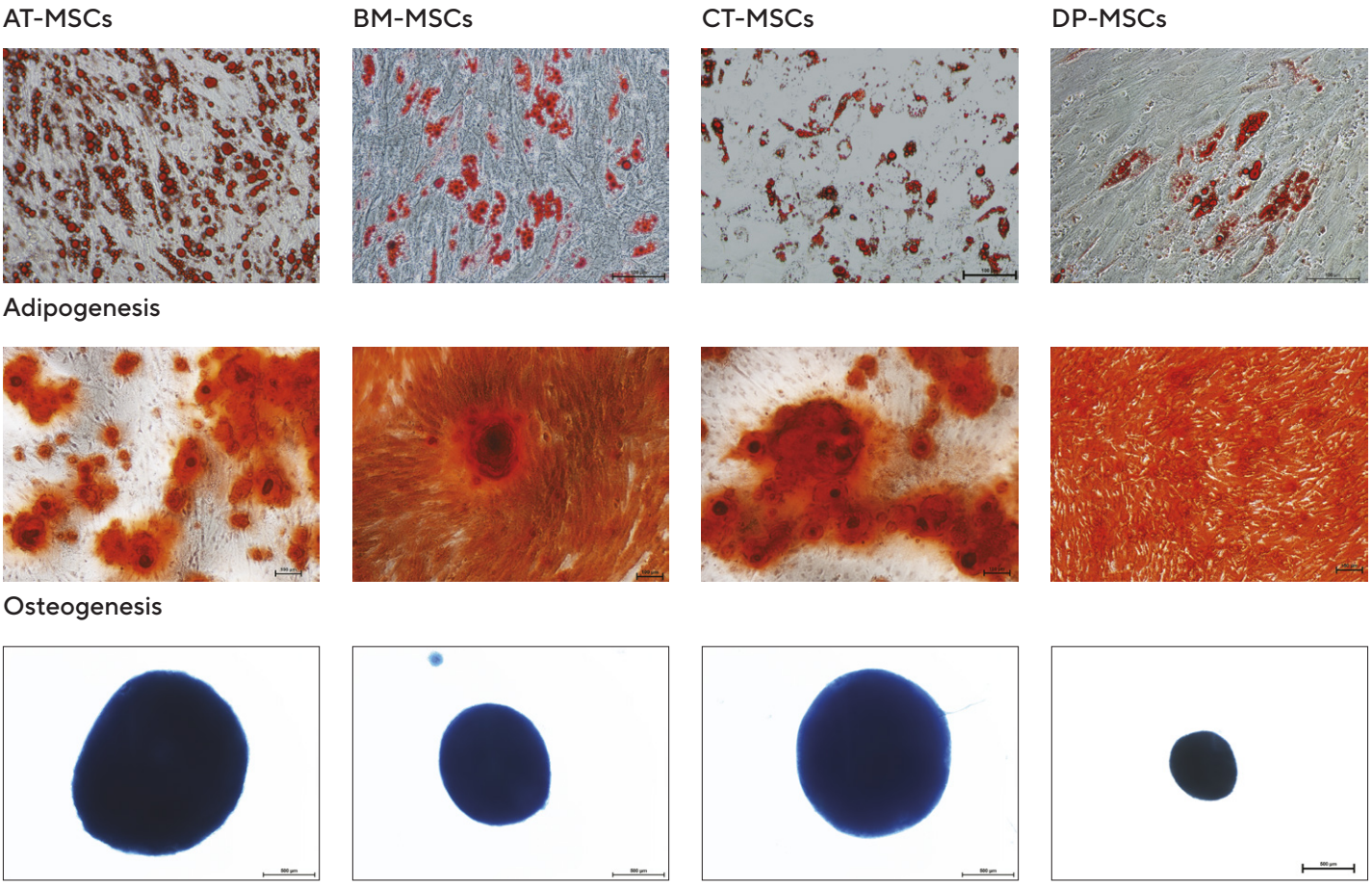
A unique line of serum-free and xeno-free differentiation media providing the ability to efficiently differentiate hMSCs from various sources (AT-MSCs, BM-MSCs, CT-MSCs and DP-MSCs) into adipocytes, chondrocytes and osteocytes.

Advantages

- **Serum-free, xeno-free**
  Eliminating the drawbacks of unwanted background differentiation and interruption in cell metabolism
- **User friendly**
  All necessary ingredients are included
- **Suitable for various sources of hMSCs**

hMSC Differentiation

Figure 30:
hMSCs from various sources were pre-cultured in MSC NutriStem XF medium and reseeded into differentiation assays using each MSCgo™ differentiation medium respectively. Representative images of 16 days assay of Adipogenesis followed by Oil red O staining (×20), 11 days assay of osteogenesis followed by 2% ARS staining (×10) and 21 days assay of Chondrogenesis followed by Alcian blue staining (×4).



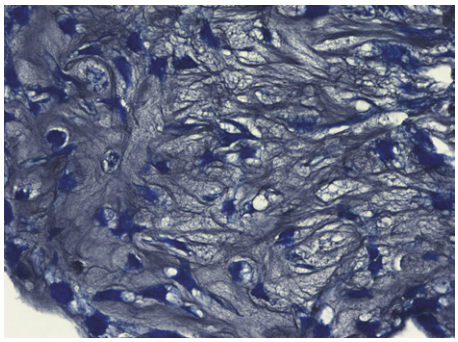
Chondrogenic Differentiation

An innovative serum-free, xeno-free medium for the initial differentiation of hMSC from various sources into chondrocytes.

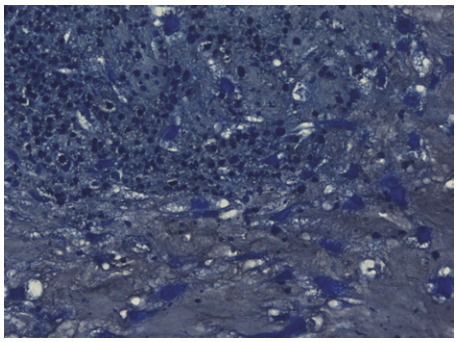
Product Name	Cat. #	Storage
MSCgo™ Chondrogenic XF Basal Medium	05-220-1	2-8 °C
MSCgo™ Chondrogenic XF Supplement Mix	05-221-1	-20 °C

Chondrogenic Evaluation

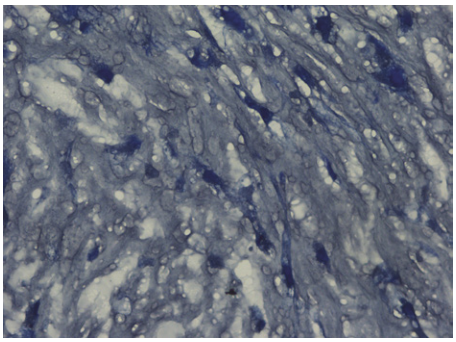
Figure 31:
Representative histological images (×40) of differentiated samples stained with Toluidine blue. Mature differentiated cells (chondrocytes) surrounded by a cartilage matrix are observed in the 3 types of hMSCs after a 21-day differentiation assay using MSCgo™ Chondrogenic XF.



AT-MSCs



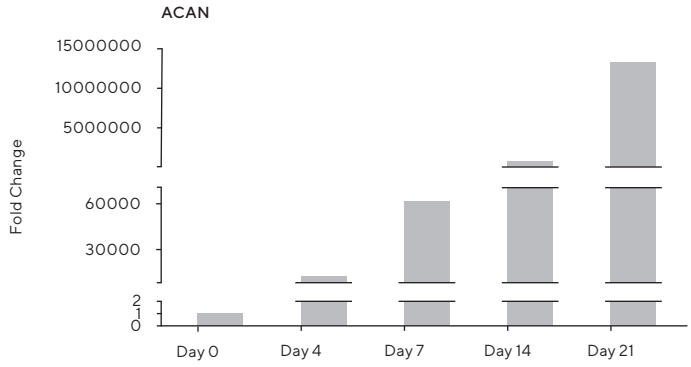
BM-MSCs

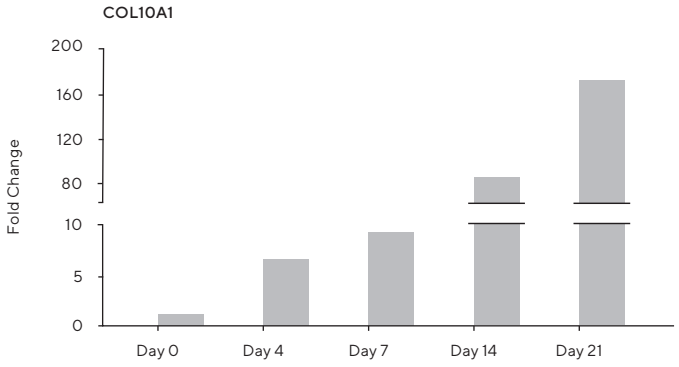


CT-MSCs

Profile Marker Expression

Figure 32:
Relative expression (RT-PCR) of chondrocytes markers during 21 days of AT-MSC differentiation assay using MSCgo™ Chondrogenic medium. Elevated expression of the chondrocyte-related genes, aggrecan (ACAN) and alpha chain of type X collagen (COL10A1), is observed.





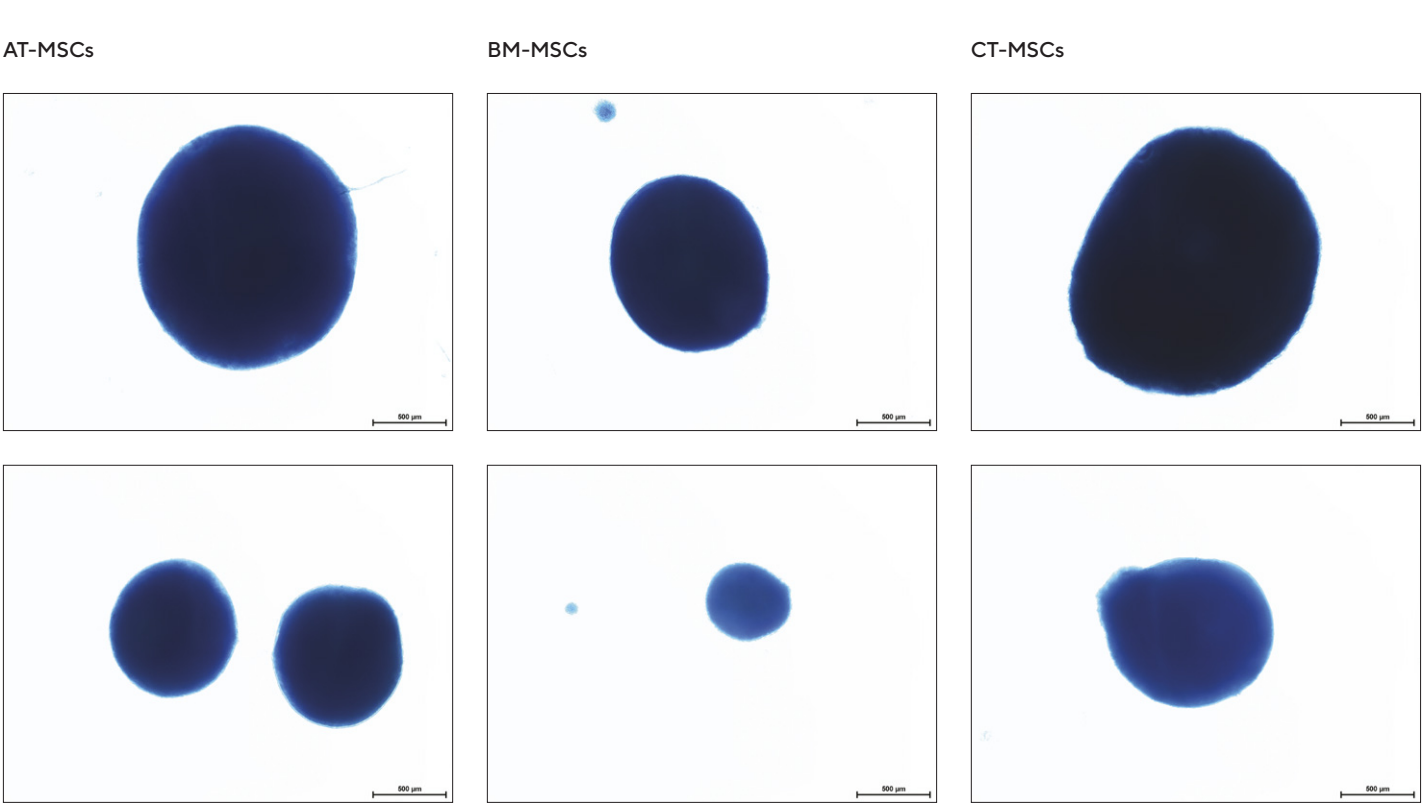
MSCgo™ Chondrogenic XF in Comparison to Other Serum-Free and Serum-Supplemented Media

Superior chondrogenesis is achieved using MSCgo™ Chondrogenic XF.
In all hMSC sources, MSCgo™ Chondrogenic XF exhibits larger cartilage spheroids with higher intensity of Alcian blue staining in comparison to other commercial SF medium.

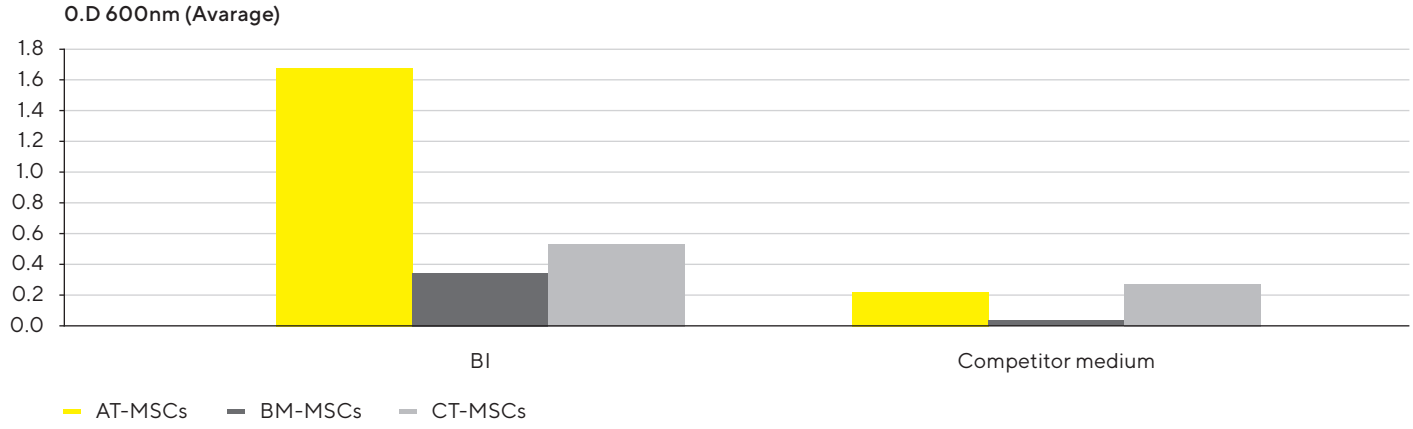
Figure 33:
Cartilage differentiation results of hMSC from various sources after 21 day assay using MSCgo™ Chondrogenic XF vs. other commercial differentiation medium, followed by Alcian blue staining.

A. And O|N elution with GuHCL (600nm)
B. The results are average of absorbance read of each well with and without the cartilage.

A



B



Adipogenic Differentiation

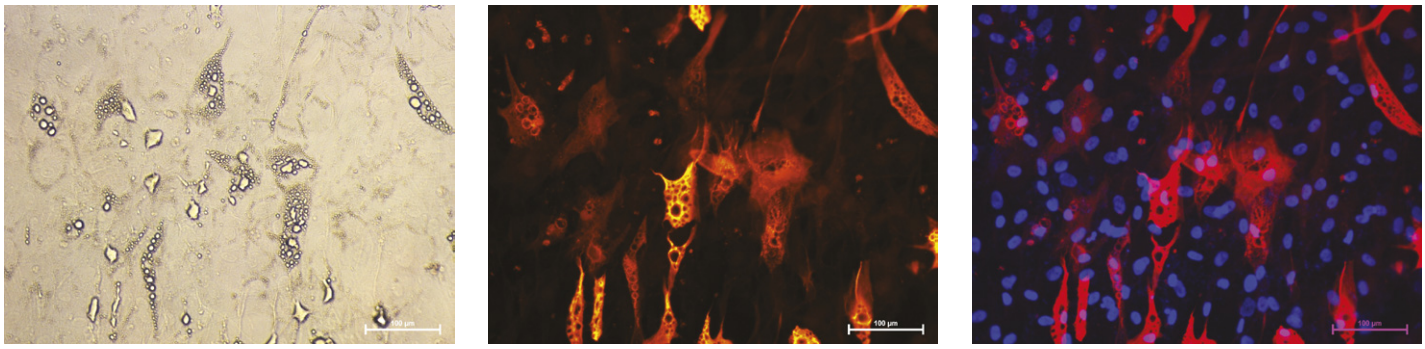
An innovative serum-free, xeno-free medium for the differentiation of hMSC into adipocytes.

Product Name	Cat. #	Storage
MSCgo™ Adipogenic XF Basal Medium	05-330-1	2-8 °C
MSCgo™ Adipogenic XF Supplement Mix I	05-331-1-01	-20 °C
MSCgo™ Adipogenic XF Supplement Mix II	05-332-1-15	-20 °C

Adipogenic Evaluation

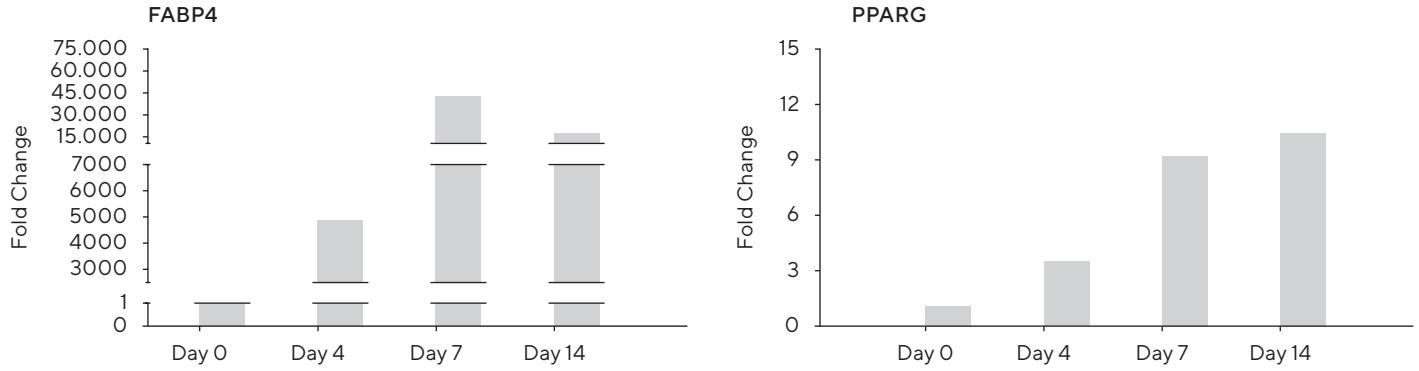
Figure 33:
Typical expression of FABP4 is observed post 11 days adipogenesis of hMSC using MSCgo™ Adipogenic XF Medium.

Light | **FABP4** | **Merged (Dapi)**



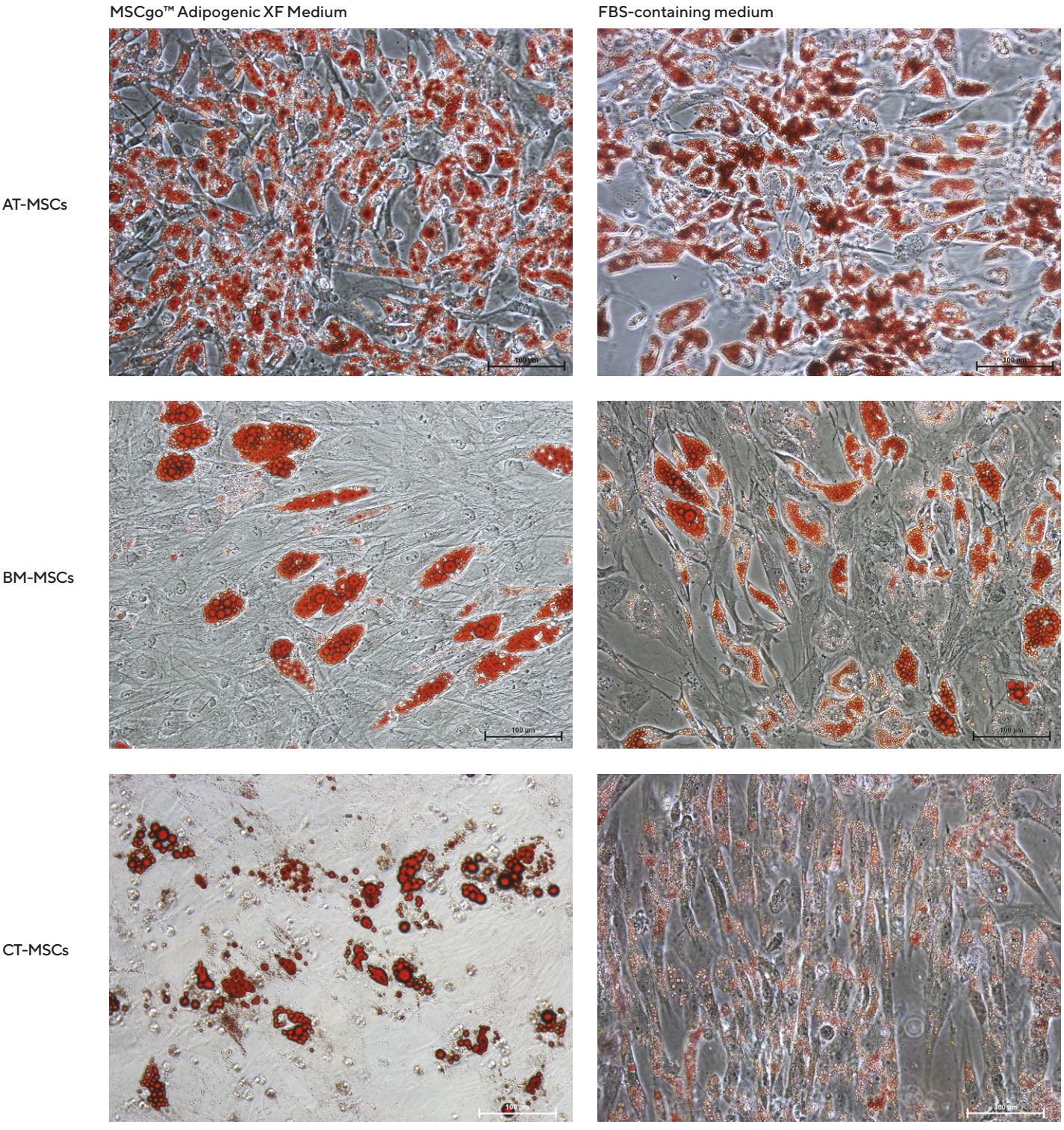
BM-MSCs

Figure 34:
Elevate expression of the adipocyte-related genes, (FABP4) and alpha chain of type X collagen (PPARG), is observed during 14 days adipogenesis of hMSC using MSCgo™ Adipogenic XF.



MSCgo™ Adipogenic XF Medium in Comparison to Other Serum-Free and Serum-Supplemented Media

Figure 35:
hMSC from various sources were differentiated into adipocytes using MSCgo™ Adipogenic XF Medium followed by Oil red O staining. MSCgo™ Adipogenic XF Medium led to similar or superior (CT-MSCs) adipogenesis in comparison to commercial FBS-containing medium (11-17 day assay).



Osteogenic Differentiation

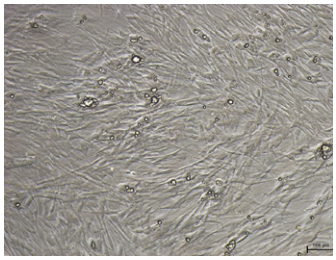
Complete, ready-to-use, xeno-free and serum-free media for the differentiation of hMSC from various sources into osteocytes.

Product Name	Cat. #	Storage
MSCgo™ Osteogenic Differentiation Medium	05-440-1	2-8 °C
MSCgo™ rapid Osteogenic Differentiation medium	05-442-1	2-8 °C

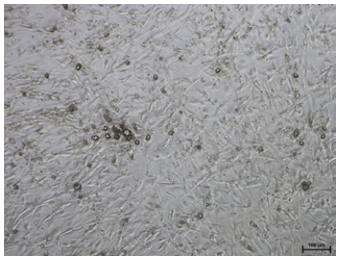
MSCgo™ rapid Osteogenic Differentiation medium will lead to faster osteogenesis (less than 10 days) in comparison to the MSCgo™ Osteogenic Differentiation Medium (10-21 days).

Osteogenic Evaluation

Figure 36:
Calcified nodules observed using both BM-MSCs and AT-MSCs after a 10 day MSC differentiation assay using MSCgo™ Osteogenic Differentiation Medium.

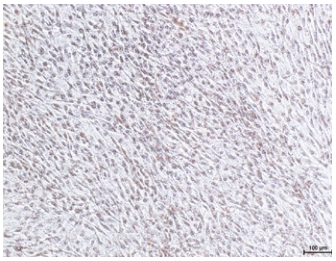


BM-MSCs

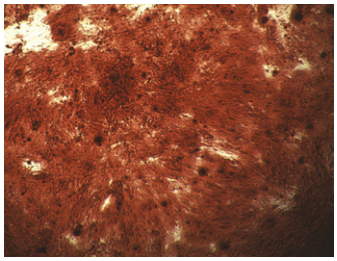


AT-MSCs

Figure 38:
Positive Alizarin staining is observed, indicates of mature osteocytes after a 28 day differentiation assay of AT-MSCs using MSCgo™ Osteogenic Differentiation Medium.

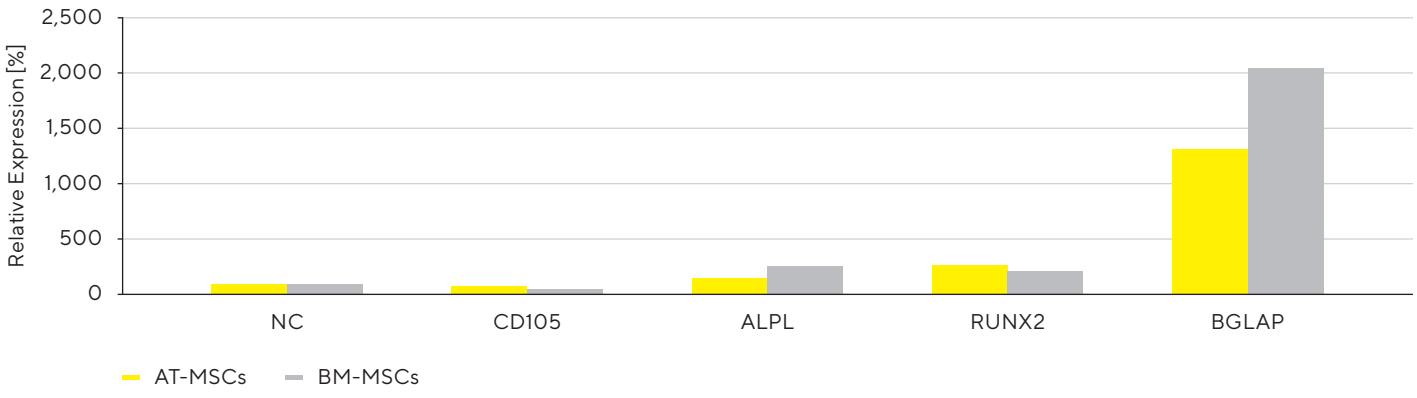


Non-differentiated cells



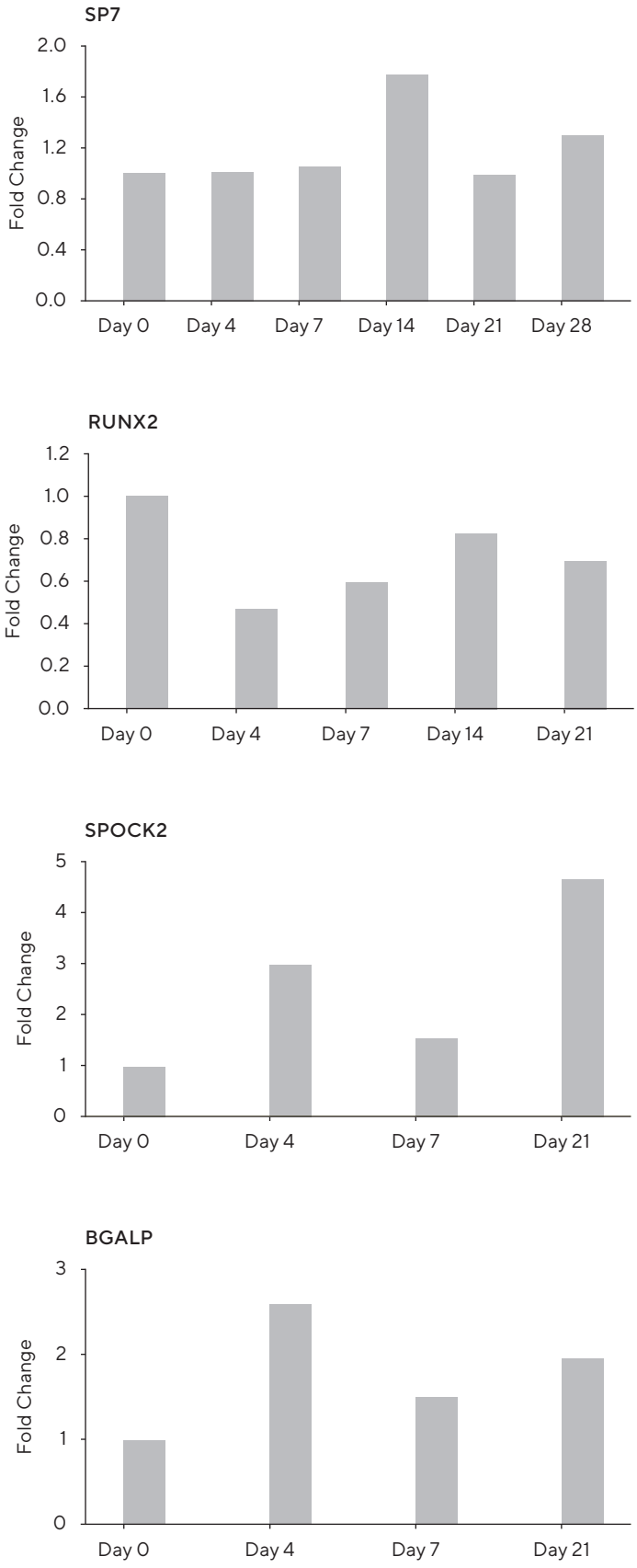
Osteogenic differentiation

Figure 37:
Relative expression (RT-PCR) of osteocyte markers after 10 days of osteogenesis of hMSC using MSCgo™ Osteogenic Differentiation Medium. Osteogenic markers were upregulated whereas an undifferentiated hMSC marker (CD-105) was downregulated. BGLAP represents a maturation state of osteogenesis.



Profile Marker Expression

Figure 39:
Profile marker expression after 28 days osteogenesis assay of hMSCs using MSCgo™ Osteogenic Differentiation Medium. Relative typical expression of the osteocyte-related genes is observed.

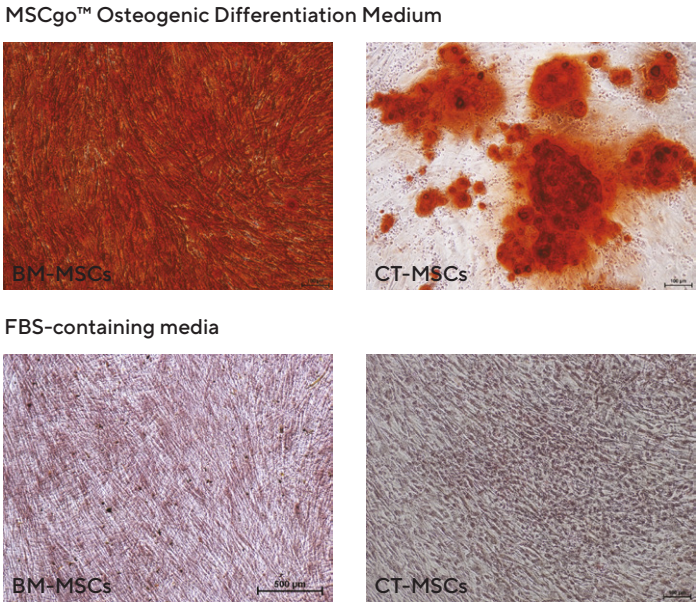


MSCgo™ Osteogenic Differentiation Medium in Comparison to Other Serum-Free and Serum-Supplemented Media

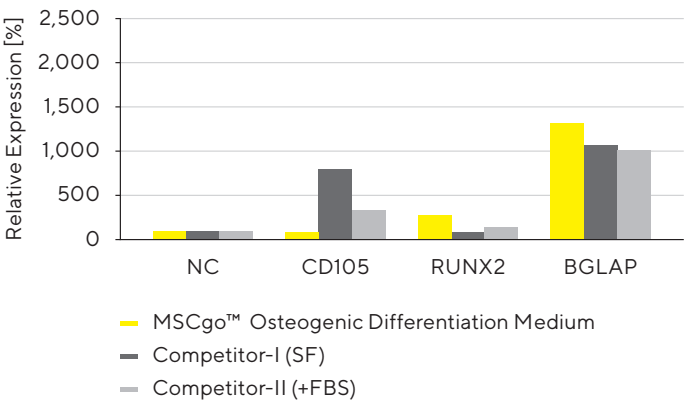
Superior osteogenesis is achieved using MSCgo™ Osteogenic Differentiation Medium.
Commercial osteogenic media are not optimal for various sources of hMSCs.

Figure 40:
A. Positive Alizarin staining after a 10 day differentiation assay is observed only when using MSCgo™ Osteogenic Differentiation Medium. **B.** MSCgo™ Osteogenic Differentiation Medium led to highest expression of osteogenic markers and lowest expression of un-differentiated hMSC marker (CD-105) in comparison to commercial media. BGLAP represents a maturation state of osteogenesis.

A



B



Ordering Information

Product Name	Cat. #	Size	Storage
MSC NutriStem® XF Basal Medium	05-200-1A 05-200-1B	500ml 100ml	2-8 °C
MSC NutriStem® XF Supplement Mix	05-201-1U 05-201-1-06	3ml 0.6ml	-20 °C
MSC NutriStem® XF Phenol Red-Free	05-202-1A	500ml	2-8 °C
MSC Attachment Solution	05-752-1F 05-752-1H	1ml 5ml	2-8 °C 2-8 °C
NutriCoat™ Attachment Solution	05-760-1-15	1.5ml	15-30 °C
PLTGold® Human Platelet Lysate (Clinical Grade)	PLTGOLD500GMP PLTGOLD100GMP PLTGOLD27GMP	500ml 100ml 27ml	-20 °C
PLTGold® Human Platelet Lysate (Research Grade)	PLTGOLD500R PLTGOLD100R PLTGOLD27R	500ml 100ml 27ml	-20 °C
Recombinant Trypsin Solution	03-078-1A 03-078-1B	500ml 100ml	15-30 °C
Recombinant Trypsin Solution with EDTA	03-079-1A 03-079-1B	500ml 100ml	15-30 °C
NutriFreez® D10 Cryopreservation Medium	05-713-1A 05-713-1B 05-713-1C 05-713-1D 05-713-1E	500ml 100ml 20ml 10ml 50ml	2-8 °C
NutriFreez® D10 Cryopreservation Medium, w|o phenol red	05-714-1A 05-714-1B 05-714-1C 05-714-1D 05-714-1E	500 ml 100 ml 20 ml 10 ml 50 ml	2-8 °C
MSCgo™ Osteogenic Differentiation Medium	05-440-1A	500ml	2-8 °C
MSCgo™ rapid Osteogenic Differentiation medium	05-442-1A 05-442-1B	500ml 100ml	2-8 °C
MSCgo™ Chondrogenic Differentiation Medium	05-220-1A	500ml	2-8 °C
MSCgo™ Chondrogenic Differentiation Medium Supplement Mix	05-221-1D	10ml	-20 °C
MSCgo™ Adipogenic Differentiation Medium	05-330-1A	500ml	2-8 °C
MSCgo™ Adipogenic Differentiation Medium Supplement Mix I	05-331-1-01	0.1ml	-20 °C
MSCgo™ Adipogenic Differentiation Medium Supplement Mix II	05-332-1-15	1.5ml	-20 °C

Sartorius and Biological Industries

Biological Industries (BI) is part of the Sartorius group. Based in Israel, we have been committed for 40 years to provide optimal and innovative solutions for cell culture practice. We manufacture and supply life science products to biopharmaceutical, academic, and government research facilities, as well as to biopharma companies.

Our diverse portfolio of products and services includes:

- Liquid and powdered cell culture media
- Novel serum-free and animal component-free media and supplements
- Products for stem cell research and cell-based therapies
- Products for mycoplasma detection and treatment
- Disinfectants
- Products for molecular biology
- Custom formulations and contract manufacturing services

All our products are manufactured via a quality management system ISO 9001:2015 and in regards to medical devices ISO 13485:2016. All aspects of the product's life cycle fall under the QMS procedures. The set-up of clean zone and clean room facilities for manufacturing are following ISO 14644, whereas the production rooms are ISO 8, storage of sterile accessories ISO 7, and filling rooms ISO 5. Aseptic filling and validation are performed according to ISO 13408.

From the outset, our policy has been based on the need to maintain an active Research and Development program in all facets of company activities. The company has its own in-house R&D department, and in addition, maintains active contact with science-based companies and research institutions in Israel and abroad, including know-how agreements with several such institutions. These ongoing efforts have led to the introduction of a series of serum-free medium products, as well as many other products for cell culture and molecular biology.

Key References

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