


MesenPRO RS™ Medium

Catalog Number 12746-012

Pub. No. MAN0007375 Rev. 2.0

 **WARNING!** Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from [thermofisher.com/support](https://www.thermofisher.com/support).

Product description

MesenPRO RS™ Medium is a reduced serum (2%) medium specifically formulated to support the growth of human mesenchymal stem cells (MSCs) in culture. MesenPRO RS™ Medium consistently improves MSC expansion compared with conventional serum-supplemented media while maintaining a comparable gene expression profile. Using MesenPRO RS™ Medium, MSCs can be expanded for multiple passages while maintaining their multipotential phenotype (i.e., differentiation into osteogenic, chondrogenic, and adipogenic lineages).

Contents and storage

Table 1 MesenPRO RS™ Medium (1 kit) [Cat. No. 12746-012]

Contents ^[1]	Cat. No.	Amount	Storage	Shelf life ^[2]
MesenPRO RS™ Basal Medium	12747-010	500 mL	2°C to 8°C; Protect from light	24 months
MesenPRO RS™ Growth Supplement	12748-018	10 mL	-20°C to -5°C; Protect from light	

^[1] MesenPRO RS™ Medium is sold as a complete kit, components are not sold separately.

^[2] Shelf life duration is determined from Date of Manufacture.

Culture conditions

Media: Complete MesenPRO RS™ Medium.

Cells: Human MSCs.

Culture type: Adherent

Culture vessels: T-flasks

Temperature range: 36°C to 38°C

Incubator atmosphere: Humidified atmosphere of 4–6% CO₂ in air. Ensure proper gas exchange and minimize exposure of cultures to light.

Procedural guidelines

- Thaw MesenPRO RS™ Growth Supplement at 2°C to 8°C, use immediately once thawed.
- Avoid repeated freeze/thaw cycles of MesenPRO RS™ Growth Supplement.
- Supplement MesenPRO RS™ Basal Medium with MesenPRO RS™ Growth Supplement, and L-glutamine or GlutaMAX™ I Supplement prior to use.
- Once supplemented, the complete MesenPRO RS™ Medium is stable for up to two weeks when stored at 2°C to 8°C protected from light. We do not recommend using beyond two weeks.

- MesenPRO RS™ Medium has been developed for the culturing of human mesenchymal stem cells that have been initiated using standard adherent isolation and growth conditions (e.g., DMEM + 10% FBS) and has not been tested for the initial expansion of MSCs directly from primary tissue sources.
- MesenPRO RS™ Medium has been developed for culturing MSCs at greater than clonal densities.
- It is recommended to subculture human mesenchymal stem cells directly into complete MesenPRO RS™ Medium.

Recover Cryopreserved MSCs

We recommend that you recover MSCs into conventional serum-supplemented medium (e.g., DMEM with 10% MSC Qualified FBS) from thaw, then subculture cells directly into complete MesenPRO RS™ Medium.

1. Rapidly thaw (<1 minute) frozen cells in a 37°C water bath.
2. Pipet the entire contents of the cryovial into a sterile 50-mL conical tube.
3. Carefully, by dropwise addition (2–3 drops per 10 seconds), add 5–7 mL of prewarmed complete conventional serum supplemented media while gently swirling the tube.
4. Add a further 5 mL prewarmed complete conventional serum-supplemented media while gently swirling the tube.

5. Transfer all the contents of the conical tube into an appropriate tissue culture flask.
6. Incubate at 37°C in a humidified atmosphere of 5% CO₂ in air.
7. Exchange spent medium with fresh prewarmed complete conventional serum-supplemented media 24 hours post-thaw.

Note: For recovery of MSCs, it is recommended to seed cells at $\geq 7 \times 10^3$ cells/cm² for the initial recovery passage.

Subculture MSCs

IMPORTANT! It is critical that cell confluency be 60–80%, cell viability be at least 90%, and the growth rate be in mid-logarithmic phase prior to sub culturing.

The following procedures apply to adherent cultures in a T-75 culture flask (75 cm²). Volumes should be adjusted accordingly for desired vessel size.

1. Observe culture flask on inverted microscope and confirm that the cells are ready to be subcultured (60–80% confluent).
2. Aspirate medium and floating cells from a confluent monolayer and discard.
3. Wash cells with 5–10 mL pre-warmed DPBS, no calcium, no magnesium, no phenol red.
4. Remove DPBS and add 5–7 mL of prewarmed TrypLE™ Express to the culture flask.
5. Incubate at 37°C until cells have fully detached (approximately 3–5 minutes).
6. Observe cell monolayer using an inverted microscope to ensure complete detachment from the surface of the flask.
7. Stop cell dissociation by adding 10 mL of prewarmed complete MesenPRO RS™ Medium to flask.
8. Gently pipet up and down to disperse cells into a single-cell suspension.
9. Transfer cell suspension into a sterile conical tube. Wash flask with an additional 5 mL prewarmed complete MesenPRO RS™ Medium and combine into the conical tube.
10. Centrifuge cell suspension at 100 × g for 5–10 minutes.










11. Aspirate supernatant and resuspend the pellet in an appropriate volume of prewarmed complete MesenPRO RS™ Medium.
12. Determine total viable cell density using a Countess™ II Automated Cell Counter (alternative automated or manual methods may be used).
13. Inoculate flask at $3\text{--}5 \times 10^3$ viable cells/cm² and return to incubator.
For optimal performance and cell growth, cultures should be re-fed every 3–4 days with fresh complete medium.

Related products

Unless otherwise indicated, all materials are available through thermofisher.com.

Item	Source
GlutaMAX™ Supplement 200 mM (100X)	35030061
L-Glutamine (200 mM) (100X)	25030081
DPBS, no calcium, no magnesium	14190144
DMEM/F-12, GlutaMAX™ Supplement	10565018
DMEM, low glucose, pyruvate, no glutamine, no phenol red	11054001
Fetal Bovine Serum, mesenchymal stem cell-qualified, USDA-approved regions	12662011
TrypLE™ Express Enzyme (1X), no phenol red	12604013
StemPro™ Accutase™ Cell Dissociation Reagent	A1110501
CTS™ StemPro™ MSC SFM	A10332-01
StemPro™ MSC SFM XenoFree	A10675-01
StemPro™ Adipogenesis Differentiation Kit	A10070-01
StemPro™ Chondrogenesis Differentiation Kit	A1007101
StemPro™ Osteogenesis Differentiation Kit	A1007201
StemPro™ Human Adipose-Derived Stem Cell Kit	R778810
Gentamicin (50 mg/mL)	15750060
Trypan Blue Solution, 0.4%	15250061
Countess™ II Automated Cell Counter	AMQAX1000

Explanation of symbols and warnings

				
Temperature Limitation	Manufacturer	Batch Code	Use By	Catalog Number
				
Caution, consult accompanying documents	Consult instructions for use	Keep away from light	Sterilized using aseptic processing techniques	

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at www.thermofisher.com/us/en/home/global/terms-and-conditions.html. If you have any questions, please contact Life Technologies at www.thermofisher.com/support.



Manufacturer: Life Technologies Corporation | 3175 Staley Road | Grand Island, NY 14072

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