

CTS™ LV-MAX™ Production Medium

Catalog Numbers A4124001, A4124002, A4124003, and A4124004

Pub. No. MAN0018636 Rev. B.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

CTS™ LV-MAX™ Production Medium is a complete, chemically defined, serum-free, and protein-free medium developed for growth and transfection of CTS™ Viral Production Cells in suspension. The medium does not contain any protein, undefined lysates, or components of animal origin.

CTS™ LV-MAX™ Production Medium is formulated with GlutaMAX™ Supplement, and does not require further supplementation. The medium is not recommended for adherent 293 cell culture.

Contents and storage

Content	Cat. No.	Amount	Storage	Shelf life ^[1]
CTS™ LV-MAX™ Production Medium	A4124001	1 L	2–8°C Protect from light	12 months
	A4124004	6 × 1 L		
	A4124002	10 L		
	A4124003	20 L		

^[1] Shelf Life duration is determined from Date of Manufacture.

Culture conditions

Media: CTS™ LV-MAX™ Production Medium for culture

Cell line: CTS™ Viral Production Cells (Cat. No. [A53956](#))

Culture type: Suspension

Shake flask type: Use PETG or polycarbonate, non-baffled, vented Erlenmeyer flasks. Cultures can be scaled up in spinner flasks or bioreactors.

Temperature range: 37°C ±0.5°C

Shaker speed: For shakers with a 19-mm orbital diameter, set the shake speed to 125 ±5 rpm. For shakers with a 25-mm orbital diameter, set the shake speed to 120 ±5 rpm. For shakers with a 50-mm orbital diameter, set the shake speed to 95 ±5 rpm.

Incubator type: ≥80% humidified, 8% CO₂ atmosphere. Ensure proper gas exchange and minimize exposure of culture to light.

Prepare media

- CTS™ LV-MAX™ Production Medium contains GlutaMAX™ Supplement and it does not require further supplementation with L-glutamine or GlutaMAX™ Supplement.
- CTS™ LV-MAX™ Production Medium is sensitive to light. Use and store the medium protected from light.
- CTS™ Viral Production Cells should be thawed, recovered, and maintained in CTS™ LV-MAX™ Production Medium.

Procedural guidelines

- All solutions and equipment that come in contact with cells must be sterile.
- Thaw and transfer cells directly into a 125-mL shaker flask with 30 mL of pre-warmed 37°C CTS™ LV-MAX™ Production Medium. Centrifugation and medium change are not required when seeding the cells.
- Do not centrifuge the cells as this can lower cell viability.
- Three days post-thaw, viable cell density should be approximately 1 × 10⁶ cells/mL with ≥ 90% cell viability.
- Subculture cells when they reach a density of ~3.5 to 5.5 × 10⁶ viable cells/mL, typically every 3–4 days.

- Allow freshly thawed cells to recover in culture for 5 passages post-thaw before using cells for virus production.
- Inspect cells to ensure that round cell morphology is maintained over passages for most of the cell population with minimal cell clumping. Proper cell phenotype is critical for optimal transfection and virus production.
- Warm CTS™ LV-MAX™ Production Medium to room temperature. For a faster warmup, place your medium at 37°C for no longer than 20 minutes.
- We recommend discarding cells after passage 20 from thaw.
- The following table shows the recommended cell density for routine cell culture.

Table 1

Subculture	Seeding density
3 day	0.55×10^6 viable cells/mL
4 day	0.35×10^6 viable cells/mL

- The following table shows the recommended shaker speed and culture volume for cultivation.

Table 2

Production vessel	Shaker speed (orbit diameter)	Vessel size	Cell culture volume range (24%–40% of vessel size)
Shaker flask (vented, not baffled)	125 rpm (19 mm)	125 mL	30 mL–50 mL
	120 rpm (25 mm)	250 mL	60 mL–100 mL
	95 rpm (50 mm)	1 L	240 mL–400 mL

Thaw and establish CTS™ Viral Production Cells

Week 1: P0–P1

1. **Day 1** Passage 0 (P0): Partially thaw a vial of CTS™ Viral Production Cells in a 37°C water bath until only a small piece of ice remains (~2–3 minutes).

Note: We recommend starting on a Monday.

2. Transfer the contents of the vial into a 125-mL shaker flask containing 30 mL of pre-warmed CTS™ LV-MAX™ Production Medium.

Note: Centrifugation and medium change are not required for these cells.

3. Place the flask on an orbital shaker platform in an incubator set to 37°C, 8% CO₂, and 80% relative humidity (for recommended shaker speeds, see Table 2).

4. **Day 4** Passage 1 (P1): Seed cells at a density of 0.35×10^6 cells/mL in a total culture volume of 40 mL in a new 125-mL shaker flask.

Week 2: P2–P3

5. **Day 8** Passage 2 (P2): Seed cells at a density of 0.35×10^6 cells/mL in a total culture volume of 40 mL in a new 125-mL shaker flask.

Note: By Passage 2, cell viability should be ≥90%.

6. **Day 12** Passage 3 (P3): Seed cells at a density of 0.55×10^6 cells/mL in a total culture volume of 40 mL in a new 125-mL shaker flask.

Note: Aliquot some cells for long-term storage.

Week 3: P4–P5

7. **Day 15** Passage 4 (P4): Seed cells at a density of 0.35×10^6 cells/mL in a total culture volume of 75 mL in a new 250-mL shaker flask.

8. **Day 19** Passage 5 (P5): Seed cells at a density of 0.55×10^6 cells/mL in 300 mL of total culture volume in a 1-L shaker flask to ensure appropriate cell numbers for lentivirus production.

Week 4: Cells are established and ready for lentivirus production

9. **Day 22** Continue to subculture cells when required until P20.

Proceed with lentiviral production.

For detailed instructions on lentiviral production, refer to the CTS™ LV-MAX™ *Lentiviral Production System User Guide* (Pub. No. MAN0018450) at thermofisher.com.

Limited product warranty

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Revision history: Pub. No. MAN0018636

Revision	Date	Description
B.0	24 December 2022	Updated Cat. No. for CTS™ Viral Production Cells.
A.0	12 April 2019	New document for CTS™ LV-MAX™ Production Medium.

The information in this guide is subject to change without notice.

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