


Efficient-Pro™ AGT™ Medium

Catalog Numbers A5322301, A5322302, A5322303, A5322304

Pub. No. MAN0025946 Rev. 1.0

 **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

Gibco™ Efficient-Pro™ AGT™ Medium is specifically designed to offer the highest batch and fed-batch culture performance and yield with recombinant CHO™ cells in a chemically defined environment. Designed to be paired with Efficient-Pro™ Feed 1 for CHO-K1 cell lines and Efficient-Pro™ Feed 2 for CHO-S and DG-44 cell lines, the chemically defined, protein-free, animal origin component-free Efficient-Pro™ AGT™ Medium provides the power to achieve high titers, high specific productivity, high protein quality, and the simplicity to start process development faster and transfer to manufacturing scale with ease.

Contents and storage

| Product | Cat. No. | Amount | Storage | Shelf Life ^[1] |
|----------------------------|----------|--------|--------------------------------|---------------------------|
| Efficient-Pro™ AGT™ Medium | A5322301 | 1 L | 2°C to 8°C; Protect from light | 12 months |
| | A5322302 | 10 L | | |
| | A5322303 | 100 L | | |
| | A5322304 | 500 L | | |

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

Culture conditions

- **Media:** Efficient-Pro™ AGT™ Medium
- **Cell line:** CHO™ cells
- **Culture type:** Suspension
- **Temperature range:** 36°C to 38°C
- **Incubation atmosphere:** Humidified atmosphere of 8% CO₂ in air. Ensure that proper gas exchange is achieved in culture vessels and minimize exposure of cultures to light.
- **Culture vessels:** Shake flasks, spinner bottles (rpm may vary with shaker platform/impeller design and should be empirically determined for optimal cell growth), or bioreactor.

Reconstitute Efficient-Pro™ AGT™ Medium

1. Measure 90% of the final volume deionized or distilled water at room temperature (15°C to 30°C).
2. Add Efficient-Pro™ AGT™ Medium at 27.3 grams/L to water.
3. Mix for a minimum of 20 minutes.
4. Use a calibrated vessel to dilute to final production volume with ambient deionized or distilled water.

5. Mix for an additional 20 minutes.
6. Measure the pH, then check and record osmolality.
7. Sterilize immediately by membrane filtration (positive pressure recommended).
Note: Once the product is filtered, use immediately or store at 2 to 8°C for up to 6 months. Protect from light.

Prepare complete medium

1. Supplement Efficient-Pro™ AGT™ Medium with L-glutamine or GlutaMAX™ Supplement at 2–8°C mM final concentration prior to use.
Note: The recommended L-glutamine and/or GlutaMAX™ Supplement concentration is 6 mM.
2. Add 10 mL/L of HT Supplement for use in applications not requiring DHFR amplification.
3. Glucose supplementation may be required for terminal batch cultures and should be determined empirically.
4. Add Anti-Clumping Agent (1 mL/L) to the medium after transfection to reduce cell aggregation, if required.
Note: Consider reducing L-glutamine concentration for fed-batch or perfusion protocols, or if the cell line in-use is

sensitive to ammonia. Addition of a surfactant such as Pluronic™ F-68 is not required.

Recover frozen cells

1. Rapidly thaw (<1 minute) frozen cells in a 37°C water bath.
2. Transfer the entire contents of the cryovial into a 125-mL shake flask containing 30 mL pre-warmed complete Efficient-Pro™ AGT™ Medium.
3. Incubate at 37°C in a humidified atmosphere of 8% CO₂ in air on an orbital shaker platform rotating at 115–135 rpm.
4. Maintain a cell density of 0.5×10^6 – 1×10^6 viable cells/mL for the first two passages following recovery; thereafter, return to your normal maintenance schedule.

Note: Do not centrifuge the cells after thawing as they are extremely fragile upon recovery from cryopreservation.

Subculture cells

1. Determine viable cell density using a Countess™ Automated Cell Counter (alternate automated or manual methods may also be used).
2. Ensure that the cell density is $\geq 1 \times 10^6$ viable cells/mL, viability is $\geq 90\%$, and the growth rate is in mid-logarithmic phase prior to subculturing.
3. Calculate the volume of cell culture and medium necessary to seed a flask at 2×10^5 – 3×10^5 viable cells/mL in a total volume of 30 mL fresh Efficient-Pro™ AGT™ Medium per 125-mL shake flask.

Note: If cell density does not reach 1×10^6 viable cells/mL within 5 days of recovery, centrifuge cells at $100 \times g$ for 5 minutes and resuspend the cell pellet in 20–30 mL of fresh complete Efficient-Pro™ AGT™ Medium.

4. Incubate at 37°C in a humidified atmosphere of 8% CO₂ in air on an orbital shaker platform rotating at 115–135 rpm.

Related products

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

Catalog numbers that appear as links open the web pages for those products.

| Item | Source |
|----------------------------|--------------------------|
| L-Glutamine, 200 mM (100X) | 25030 |
| GlutaMAX™ Supplement | 35050 |
| HT Supplement (100X) | 11067030 |
| Anti-Clumping Agent | 0010057 |
| Efficient-Pro™ AGT™ Feed 1 | A52091 |
| Efficient-Pro™ AGT™ Feed 2 | A52216 |
| Efficient-Pro™ Feed 1 | A52088 |
| Efficient-Pro™ Feed 2 | A52214 |
| Efficient-Pro™ Medium | A53222 |

Limited product warranty

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For descriptions of symbols on product labels or product documents, go to thermofisher.com/symbols-definition.

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