

10	Document ID:	TDS-LMG-001-100ML	Version:	001
	Date of Issue:	10-JAN-2023	Approved by:	Dr. Iman Kamranfar
• SERANA Science for Life	Review Date:	10-JAN-2025	Signature:	ifter
	Title:	TEC	CHNICAL DATASHEE	Γ

#### LYMPHOGROW I

Complete Karyotyping Medium for Peripheral Blood Lymphocytes, Sterile-filtered

Eiltration Treatment	Sterile Filtered; Contains preselected serum, L-Glutamine and		
Filtration, freatment	antibiotics.		
Product Code	LMG-001-100ML		
Shelf Life	24 months from DOM		
Starrage Tananatana	Store between-5°C to -20°C protected from light. Once opened, store		
Storage Temperature	at +2°C to +8°C and use within 2 weeks		
Shipping Temperature	Frozen (Dry ice)		
Thewing	Thaw the medium at 2°C to 8°C, or alternatively at 37°C in water bath		
nawing	and swirl gently to homogenize		
CO2 concentration, optimum	5%		

#### **QC** Specifications

Physical and Chemical Analysis	Method	Specifications	Units
Appearance	Visual	Clear amber to red frozen liquid	n/a
pH at RT	Electronic pH Meter	6.8 - 7.6	n/a
Osmolality	Osmometer	Test and report	mOsm/kg
Endotoxin	LAL Kinetic	≤ 10.0	EU/ml
Sterility			
Aerobic Bacteria	EP 2.6.1	Not detected	n/a
Anaerobic Bacteria	EP 2.6.1	Not detected	n/a
Fungi (Yeast & Mold)	Internally Validated	Not detected	n/a
Functionality Test Internally Validated		Pass	Pass

#### **GENERAL INFORMATION/FORMULATION**

*LYMPHGROW I* formulation has been developed and optimized by Serana R & D team and its superiority over the commercially available similar products were approved for the cytogenetics-related parameters in short-term and long-term cultivation of Peripheral Blood Lymphocytes, which are intended for the preparation of karyograms, fluorescence *in situ* hybridization and other cytogenetic methods. The medium is supplied frozen.

#### Important information:

This medium is ready to use and no further supplements are needed. *LYMPHGROW I* is formulated based on the basal medium and already supplemented with preselected foetal bovine serum, L-Glutamine and antibiotics.

#### **INSTRUCTION FOR USE**

Lymphocytes are a subtype of leukocytes substantial for immunoregulation and play an important role in the development of promising and feasible immunotherapeutic cancer therapies. Karyotyping of peripheral blood lymphocytes (PBL's) has been proved to be an inalienable tool for characterization of the complex chromosome rearrangements in leukemia (68). The determination of leukemia specific cytogenetic abnormalities provides insight into leukemia pathogenesis and allows integral prognostic assessment.

Lymphocytes usually do not undergo subsequent cell divisions. In the presence of a mitogen, inducer of the cell division, lymphocytes are triggered to enter into mitosis. After 48 – 72 hours, colcemid is added to the culture as the mitotic inhibitor, arresting mitosis on the metaphase stage by interfering with the formation of spindle apparatus. After treatment by hypotonic solution, fixation and staining, chromosomes can be microscopically observed and evaluated for abnormalities.

1. We recommend to initially thawing *Lymphogrow I* Medium per described and making aliquots of 10 ml sterile tubes.



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2. Thaw the precalculated amount of *Lymphogrow I* per described.

3. Transfer 0.5 ml of heparinized whole blood into a tube containing 10 ml LymphoPrime Medium.

4. Incubate the culture at +37°C, 5 % CO2 in an incubator for 72 hours.

5. Add Colcemid Solution (Product Code: *CDS-002-10ML*) to each culture tube at a final concentration of  $0.1 \mu g/ml$ ; 0.1 ml in each 10ml of medium).

6. Incubate the culture for additional 30-60 minutes.

7. Transfer the culture to a centrifuge tube and spin at 500 g for 5 minutes.

8. Remove the supernatant and resuspend the cells in 5 ml of hypotonic 0.075 M KCl (Product Code: CDS-003-100ML),

prewarmed to +37°C. The hypotonic solution allows swelling of the cells to increase the visibility of chromosomes.

9. Incubate at +37°C for 10 minutes.

10. Spin at 500 g for 5 minutes.

11. Resuspend the cell pellet in a 5 ml of an ice-cold fixative (1:3 solution of AcOH and MeOH) dropwise. The fixative kills the cells, removes cell debris and helps to preserve cellular structures.

12. Leave at + 4ºC for 10 minutes.

13. Repeat steps 10-12.

14. Spin at 500 g for 5 minutes and discard the supernatant.

15. Resuspend the cell pellet in a small volume (0.5 – 1 ml) of fresh fixative, drop onto a clean slide and allow to air dry.

16. stain the cells with Orecin or Giemsa using protocol established in your laboratory.

#### PRECAUTIONS AND DISCLAIMER

The medium is not intended for therapeutic use.

Each laboratory is obliged to perform representative tests according to the valid legal regulations and in its own environment to ensure that it is suitable for this purpose before the medium can be used in routine diagnostics.

Do not use if a visible precipitate is observed in the medium.

Do not use this medium beyond the expiration date indicated on the product label.

India Contact:

Life Technologies (India) Pvt. Ltd. 306, Aggarwal City Mall, Opposite M2K Pitampura, Delhi – 110034 (INDIA). Ph: +91-11-42208000, 42208111, 42208222, Mobile: +91-9810521400, Fax: +91-11-42208444 Email: customerservice@lifetechindia.com Website: www.lifetechindia.com





## Serana Europe - Leading Manufacturer and Supplier of Cell Culture Products

Serana Europe GmbH is a leading manufacturer and supplier of cell culture products. Our product range includes animal & human sera, sterile liquid & powdered classical media, reagents, supplements, and buffer solutions for cell culture applications. Serana's products are used in all areas where cell culture is performed. This includes the biopharmaceutical industry for the production of vaccines, therapeutic proteins and diagnostics. In addition, we are a major supplier to Academic R&D institutes (universities, hospitals & clinics), private research organizations and various biotech companies.



We are EDQM, ISO 9001, and ISO 13485 certified

#### Our quality promise

Products manufactured by Serana utilize stringent operating and quality control procedures. Detailed production and traceability records are available for every batch produced. A battery of QC tests are performed including: physical and chemical analysis, protein profiling, sterility, virology, biochemistry and cell culture performance benchmarking. Only batches that pass Serana's rigorous quality control procedures are released for sale. Detailed certificates of analysis are prepared and made available for each lot produced.

Our quality assurance team guarantees exceptional quality manufacturing standards which follows relevant guidelines and regulatory requirements.We are EDQM, ISO 9001, and ISO 13485 certified, and our team is dedicated to continuously improve our quality management system. Our quality control department also offers a wide variety of relevant biological, molecular and biochemical assays as a service. We have certified manufacturing facilities in Germany and Australia, which makes us an ideal global partner with validated storage and delivery logisitics world-wide.

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Dorfstraße 17A 14641 Pessin Brandenburg Germany India Contact: Life Technologies (India) Pvt. Ltd. Ph: +91-11-42208000, 42208111, 42208222 Mobile: +91-9810521400 Email: customerservice@lifetechindia.com www.lifetechindia.com



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### DIAGNOSTIC PRODUCTS CYTOGENETICS AND SUPPLEMENTS

# LMG-001

## Lymphogrow<sup>TM</sup>

## Complete and superior Karyotyping Medium for Peripheral Blood Lymphocytes

#### **Advantages**

- Optimized superior formulation by Serana R & D team
- Superior chromosomal morphology (Fig. 1) and higher mitotic index (Fig. 2)
- Superior growth support of short-term and long-term lymphocyte cultivation
- Consistent Lot-to-lot performance
- Comprehensive QC specifications (in product TDS)
- Manufactured at Serana ISO 13485-certified facility in

#### Convenience

- Complete, ready-to-use medium Fully supplemented with FBS, antibiotics, and L-glutamine
- Detailed available IFU in the product Technical datasheet.
- Flexible to be ordered in different volume packaging from 50ml to 1000ml per Unit.



Fig. 2. Superiority of LymphoGrow™ in providing higher Mitotic Index (MI). MI ± SE (0.5 h colcemid incubation time). One-way ANO-

VA with subsequent Tukey HSD post hoc test (p < 0.001).





Fig 1. Mitotic spreads of human PBL's cultured with LymphoGrow™ as described in the product TDS.

Photomicrograph of a microscope slide stained with Giemsa. Left: Sister chromatids during prophase; right: Chromosomes during Prometaphase.



Mitotic Index /1000 Cells

Fig. 3. Superiority of LymphoGrow™ in providing higher Lymphocyte growth over eight days of incubation. One-way ANOVA with subsequent Duncan post test (*p* < 0.001). Error bars represent SE.



AmnioGrow™	AMG-001
Phytohemagglutinin (PHA-M)	CDS-001
Colcemid solution	CDS-002
Potassium Chloride solution	CDS-003
Sodium Citrate Solution	CDS-004
Trypsin EDTA Solution	CDS-005

For the latest technical data and pricing, please refer to the Serana-Europe website. serana-europe.com/products/rll-001-100ml



#### ORDERING INFORMATION

PRODUCT	OPTIONS	VOLUME	STORAGE	SHELF LIFE	CODE
Lymphogrow™	Complete and superior Karyotyping Medium for Peripheral Blood Lymphocytes		-15 to -20°C	2 Years	LMG-001-100ML

For complete current specifications and other technical information please see the technical data sheet on our website www.serana-europe.com Should you have any specific product requirements that our stock product offering cannot fulfill;

please don't hesitate to ask if we can tailor-make a product to your specifications.