Gelacell™ - PLLA 24 well plate

Bringing cell culture to the next dimension

Product Description

Gelacell™ is a unique, 3D microfibrous scaffold specifically engineered for advanced in vitro 3D cell culture and tissue engineering applications. Designed as a non-woven, highly porous scaffold, Gelacell™ offers exceptional biocompatibility and non-toxicity across a variety of cell types. Our specially integrated circular scaffolds in a well plate format (as shown in Figure 1) offer a straightforward visual imaging of cells directly from the well plate.

Perfectly tailored for the cultivation of complex systems like organoids, cell microenvironments, and tissue regeneration, Gelacell™ also makes a robust platform for drug screening and other cell culture investigations. The scaffold's optimized design ensures thermal, chemical, and mechanical stability, while providing substantial swelling capabilities and porosity for efficient nutrient diffusion and cellular waste build up prevention.

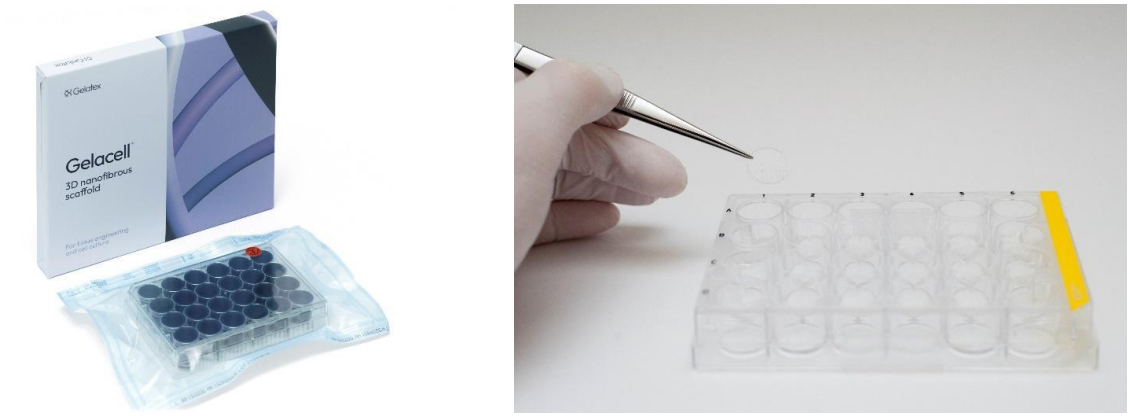

Figure 1. Gelacell™ scaffolds incorporated in the well plate.

Product Features

- 3D architecture with a substantial available surface area.
- Minimal modifications required for 2D to 3D culture transition.
- Stable mechanical properties.
- Scaffolds are affixed to transparent, inert discs for easy handling.
- High porosity promotes cell migration and efficient diffusion of nutrients, solutes, and gases.
- Compatibility with a variety of cell lines and culture conditions.
- Biocompatible polymers, diverse and widely accepted in cellular applications.
- Sterilized by Gamma irradiation and will remain sterile until the pack is opened.
- Storage at room temperature.

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Product Datasheet

Product information	Gelacell™
Product code	GC0805RN-WP24-A
Polymer	Poly-L-lactide (PLLA) (Pharmaceutical grade)
Appearance (dry)	White
Appearance (swelled in water/PBS)	White (Translucent)
Fiber orientation[1]	Random
Mean fiber diameter[1]	0.4 – 1.4 µm
Thickness	140 - 230 µm
Area density	3.0 – 9.0 g/m^2
Porosity[2]	95% - 99%
Wettability[3]	Hydrophilic The material uptakes water, and compounds that dissolve in water due to the porosity and capillary forces wherever the material is soaked in a solution and incubated.
Absorptivity in PBS (pH 7.4 at 37°C)[4]	600% - 1200% The material was kept soaked for 24 hours in PBS at 37°C
Degradation (in PBS at 37°C)[4]	Up to 20%
pH (of PBS solution after 24 hours)[4]	7.3 – 7.5 without observable changes
%Elongation[5]	20% - 45%
Elastic modulus[5]	400 – 1300 kPa
Ultimate tensile strength[5]	80 – 220 kPa

1. Information is derived from SEM images.
2. Porosity is determined by comparing the apparent densities of scaffolds to the bulk densities of PLLA.
3. Wettability is determined by visual observation when water/PBS was added dropwise on top of the scaffolds after washing the scaffolds with 70% ethanol. Ethanol washing is highly recommended as PLLA is inherently hydrophobic in nature.
4. Degradation and absorptivity tested in PBS solution. Absorptivity evaluates solution uptake, while degradation is tracked as weight reduction over time with bi-daily PBS renewal.
5. Tensile strength measured using a specific in-house protocol, conducted in dry conditions.

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Morphology of PLLA Aligned scaffold

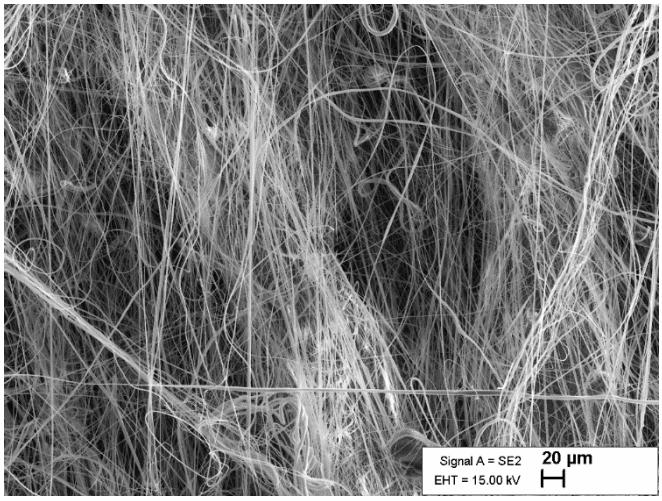

Figure 2. SEM image showing fiber morphology and porosity.

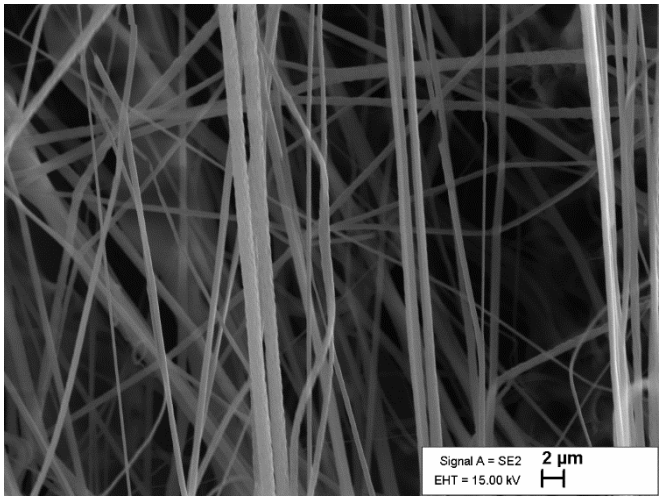

Figure 3. SEM image showing fiber diameter and distribution.

The morphology of fibrous scaffolds plays a pivotal role in its interaction with eukaryotic cells derived from humans and animals. The unique structure of microfibers shown in Figure 2 and Figure 3, mimicking the extracellular matrix, offers a high surface area, and interconnected porous network. This morphology provides an advantageous environment for cell adhesion, proliferation, and differentiation.

Biocompatibility of PLLA Scaffold

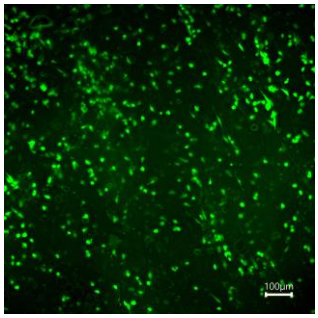

Figure 4. Hela cells after day 1.

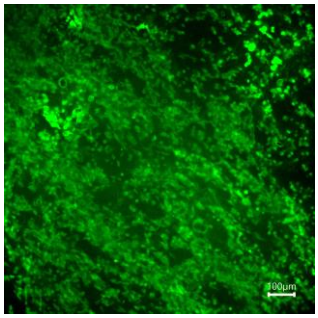

Figure 5. Hela cells after day 3.

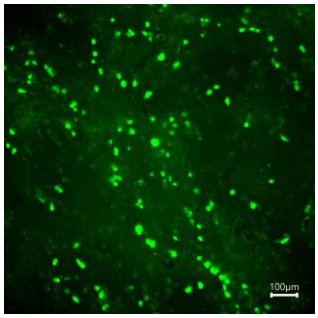

Figure 6. Huh7 cells after day 1.

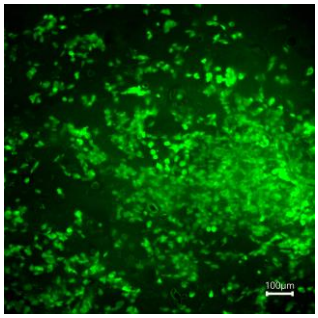

Figure 7. Huh7 cells after day 3.

The scaffolds exhibit promising biocompatibility with various cell types, including HELA, HEK293, HUH7, HEPG2, U2OS, BHK21, and C2C12. Their biomimetic design and specialized polymer composition enhance cell-material interactions, supporting key processes like cellular attachment and spreading - crucial for cell growth and function. HELA and HUH7 cell lines show adherence, proliferation, and infiltration within the scaffold structure (as depicted in Figure 4 to Figure 7). This compatibility is vital across diverse tissue engineering applications, enabling these cell lines to thrive within the scaffold's microenvironment.

Standard Operating Procedure

Gelacell unpacking and sterilizing

1. Adhering to aseptic techniques, place the Gelacell™ package under a laminar flow hood, take the well plate out of the package and remove the blue tops (Figure 8).
2. The production of Gelacell takes place in clean rooms ISO Class 7 conditions and the products are sterilized by Gamma irradiation. UV sterilization for 20 minutes is advisable to retain sterility of the product upon unpacking.

Note: The scaffolds are expected to remain unchanged under standard UV-C radiation dosage and standard gamma radiation dosage (15-25 kGy). Higher irradiation doses should be used with caution.

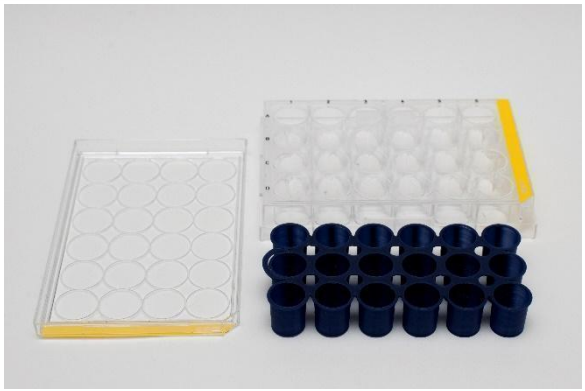

Figure 8. Unpacking Gelacell™ well plate and removing the blue tops.

Cell seeding and culture

1. Before cell seeding, wash the scaffold once with 70% ethanol, followed by a rinse with PBS solution three times.
2. Pre-swell the scaffold with your preferred cell culture media (with/without serum). Add 200 µl - 300 µl of media onto the scaffold, incubate for 30 min to 1 hour, and aspirate the spent media.
3. After pre-swelling, allow the scaffold to partially dry in a laminar hood by keeping the lid open for 20 minutes. It helps in absorbing the cell seeding volume and spreading the cells uniformly.
4. Dispense your desired concentration of cell culture suspension with complete media onto the scaffold. The recommended cell seeding density ranges from 10^4 to 10^6 cells/well and cell suspension volume ranges from 10 µl to 70 µl. However, the exact figure will depend on the cell type and the planned duration of cell culture.
5. Place the well in a CO2-incubator at 37°C for 30 minutes - 2 hours to allow initial cell adhesion. Afterward, gently fill the well with 1 ml medium without dislodging the cells that have already adhered to the scaffold. Return the scaffolds to the incubator for culturing cells.
6. It's recommended to exchange media roughly every 24 hours to 48 hours; however, this may vary depending on the cell line, media, and cell density. Aspirate the waste media from the sides of the well and carefully add fresh media on top of the scaffold without dislodging the adhered cells. Continue media exchange throughout the cell culture period.

Note: The partial drying of scaffold is a crucial step to retain most of the cells during seeding and controls the moisture content before introducing cells. The rate and efficiency of cell attachment and detachment can be affected by temperature, pH, nutrient exchange, the concentration of cells, enzymatic degradation, and cell staining. The above guidelines are the best-known practices based on the tests carried out on the scaffolds. The cell seeding volume is preferred to be in between 10 to 70 µm to achieve total cell adhesion and avoid cell losses. However, this seeding volume is not restricted for lower (< 10^4) or higher (> 10^6) seeding density, except the performance of cell adhesion might differ.

Cell culture analysis

1. **[Optional] Cell fixation:** Rinse the scaffolds with PBS and use 3.7% - 4% formaldehyde or paraformaldehyde for cell fixation. Leave the scaffolds in the solutions for 15 min at room temperature or in a CO2-incubator.
2. **For cell viability:** Use assays such as MTS, MTT, CCK-8, etc. for determining the cell activity on the scaffolds. Follow the standard protocols suggested by the manufacturer. The recommended volume of assays is 400 µl - 500 µl /well.
3. **For cell staining and imaging:** Utilize stains such as calcein, FDA, phalloidin-conjugates, DAPI, or propidium iodide, alone or combined, to enhance contrast during imaging. Cells can be observed by placing the well plate directly under a fluorescence microscope. However, for improved handling and enhanced imaging, transferring the scaffolds onto a glass slide is highly recommended. Adding a staining volume of 400 µl is advisable for optimal results.
4. **Handling:** Employ fine-point tweezers, either straight or angled, to grip the discs (refer to Figure 1). Position the discs facing the scaffolds on a glass slide that contains one or two drops of mounting media. This step is crucial to secure the discs in place. (Figure 9).
5. **Imaging:** Evaluate the scaffolds under a fluorescence or confocal microscope for imaging and visualization.

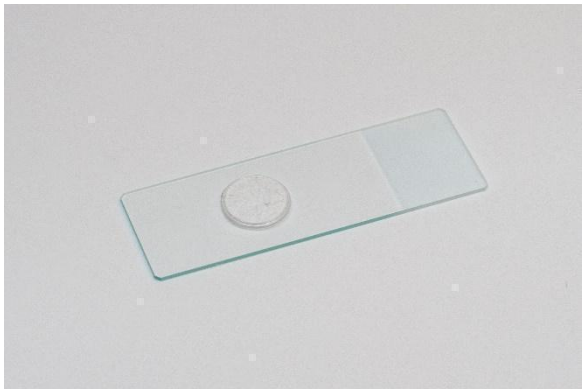

Figure 9. Fix the Gelacell™ discs using mounting media for imaging.

Note: The method of cell/tissue culture analysis on the scaffold may vary based on the tests and applications. The above guidelines are based on the tests conducted on the scaffolds. However, users are advised to extend the scope of analysis as per their requirements.

Important Information:

- The scaffold is compatible with both serum and serum-free cultures.
- The transparent discs are not fixed to the wells. Handle with care when transferring or changing solutions in the wells to prevent flipping the discs. Avoid forceful pipetting.

- Avoid scratching the scaffold discs while using micropipettes. When pipetting solutions from/into the well, place the tip at the well's periphery to avoid contact with the scaffold.
- The product maintains sterility until the package is opened, with a shelf life of 24 months from the dispatch date. Both sterility and shelf life are preserved when the product is stored according to the specifications outlined in the Safety Data Sheet.