alvetex® Frequently Asked Questions

How is alvetex® supplied?

alvetex® products are gamma irradiated and supplied in sterile, individually-wrapped blister packs.

What formats of alvetex® scaffold are available?

Alvetex® Scaffold are currently available in the following formats: 12-well plates (AMS.AVP002), 24-well plates (AMS.AVP006), 96-well plates (AMS.AVP009) and 384-well plates (AMS.AVP009). In addition, there are 6-well inserts (AMS.AVP004-32), 12-well inserts (AMS.AVP005-34) and we have recently launched perfusion plates (AMS.AVP011) for dynamic media flow and perfusion across cells culture. For more information, please refer to www.amsbio.com/alvetex.aspx or contact info@amsbio.com

What formats of alvetex® strata are available?

Alvetex® Strata are currently available in the following formats: 6-well inserts (AMS.STP005-12) and 12-well inserts (AMS.STP004-12). For more details please visit www.amsbio.com/alvetex.aspx or info@amsbio.com

Are other formats available?

We are continually optimising our current products and developing new formats. Custom made formats of Alvetex® scaffold are available for collaborations, please contact info@amsbio.com for details.

What is Alvetex® made of?

Alvetex® is a cross-linked polystyrene scaffold supplied with a thickness of 200µm. It is highly porous, inert and does not degrade.

What is the porosity of Alvetex®?

Alvetex® has a porosity of greater than 90%.

What are the differences between Alvetex® scaffold and strata?

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<th>voids diameter</th>
<th>interconnects diameter</th>
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<tbody>
<tr>
<td>scaffold</td>
<td>40 µm</td>
<td>14 µm</td>
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<tr>
<td>strata</td>
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What are the diameters of Alvetex® inserts?

6-well and 12-well inserts Alvetex® scaffold and strata are holding discs of 22 mm and 15 mm diameter respectively.

Is Alvetex® scaffold supplied sterile?

Alvetex® scaffold products have been sterilised by gamma irradiation and remain sterile until the blister packs are opened.

Can you autoclave Alvetex® scaffold?

The Alvetex® scaffold disc itself can be autoclaved, ensuring it has been removed from the insert or plate first.

Is Alvetex® scaffold reusable?

Alvetex® scaffold is a single use, disposable product.

Does Alvetex® scaffold have an expiry date?

There is no expiry or shelf life for Alvetex® scaffold. The plate or inserts are sterile and ready for use as long as the packaging seal remains intact.

What is Alvetex® proper storage conditions?

All Alvetex® products are shipped and stored at room temperature.

Is Alvetex® scaffold chemically resistant?

Owing to the cross-linked nature of the polystyrene scaffold, it is chemically resistant; however the surface properties can be affected by certain chemicals. Some solvents like acetone may swell the scaffold. Please contact technical support if you have a specific query.

Does Alvetex® scaffold autofluoresce?

No significant levels of autofluorescence have been observed from Alvetex® scaffold using standard wavelengths of light.
What is the significance of ethanol treating Alvetex® scaffold?

Alvetex® scaffold needs to be treated with ethanol in order to render it hydrophilic. If this step is missed out and cells are seeded directly onto the untreated scaffold, they sit in a droplet on the top and are unable to penetrate and grow in 3D.

Is there disc to disc variability?

Each batch of Alvetex® scaffold is tested for its physical properties, porosity and suitability for cell attachment and viability. Only batches fulfilling strict criteria are released by our ISO9001:2008 certified Quality System.

Have any papers been published regarding Alvetex® scaffold?

Numerous peer-reviewed scientific articles describe the development and application of Alvetex® scaffold, which can be accessed from www.amsbio.com/alvetex.aspx or by contacting info@amsbio.com. Scientists at Reinnervate are continuously researching novel applications for this technology, publishing and presenting data when appropriate.

Which cell types have been tested on Alvetex® scaffold?

To date we have released protocols for culturing the following cell types: 3T3, HaCaT, HepG2, Tera2.cl.SP12 and CHO-K1, which can be accessed through www.amsbio.com/alvetex.aspx. Other cells which have been successfully grown on Alvetex® scaffold include bone marrow stromal cells, primary hepatocytes, adipose tissue-derived stem cells, human pluripotent stem cell-derived neurons, MG63 osteoblast-like cells, cylindroma primary cells, SW480 colon carcinoma cells, MET4 squamous carcinoma cells, L929 mouse fibroblasts, U118-MG glioblastoma cells, MCF-7 cells and neural crest cells, among others.

Are there any protocols available for the users?

A range of example protocols can be found in www.amsbio.com/alvetex.aspx. These are being continually updated and expanded upon and currently include general information about choosing and using Alvetex® scaffold; examples of how to grow specific cell types; compatible analytical techniques and specialised applications.

How can you see your cells growing in 3D in Alvetex® scaffold?

Neutral Red dye can be used to quickly and easily monitor whether cells are attaching and proliferating using a standard light microscope. Other options to visualise cells growing in 3D in Alvetex® scaffold include, but are not limited to, live cell imaging, fluorescent marker analysis, confocal analysis, histology using a range of cytological stains and electron microscopy. Please refer to www.amsbio.com/alvetex.aspx.
Can you visualise live cells using Alvetex® scaffold?

For following culture progress, the use of dyes can be employed to visualise cells against the scaffold background using light microscopy. A full protocol for this simple method is available in www.amsbio.com/alvetex.aspx. More complex techniques that are commonly used for tissue processing can also be implemented with excellent results.

What analysis techniques are compatible with Alvetex® scaffold?

Alvetex® scaffold is compatible with a broad range of general cell and molecular techniques. These include various biochemical assays, histology (tissue processing, fixation, embedding and sectioning), fluorescence microscopy, immunochemistry, in-situ hybridisation, electron microscopy, bright field and phase microscopy; extraction of nucleic acid and total protein etc.

Can you coat Alvetex® scaffold?

Yes, most standard coating methods for cell culture plastic are compatible with Alvetex® scaffold including the use of extra-cellular matrix proteins, poly D/L lysine etc. A range of protocols are available in www.amsbio.com/alvetex.aspx and many extra-cellular matrix proteins in http://www.amsbio.com/Cells-Extracellular-Matrices.aspx.

Can you remove cells from Alvetex® scaffold?

Partial cell recovery is possible. Cells can be removed from the scaffold using a combination of trypsin and mild agitation. For an example protocol, please visit www.amsbio.com/alvetex.aspx. For most techniques such as total protein or nucleic acid isolation, cell retrieval is not required or recommended.

How do you estimate cell confluency in Alvetex® scaffold?

Simple dyes can be used to estimate cell culture confluence and viability. A full protocol for a simple method is available in www.amsbio.com/alvetex.aspx. An extensive number of end-point visualisation techniques are also compatible, including live cell imaging, fluorescent marker analysis, confocal analysis, biochemical assays, histological analysis using a range of cytological stains and electron microscopy.

Can you get reproducible data using Alvetex® scaffold?

Reproducibility of data using Alvetex® scaffold is very good. Where the same cell suspension has been used, the same growth pattern for the cells is consistently obtained. It should be noted that some cell types prefer to occupy the top quarter of the scaffold, whereas others invade all the way through.
Can you explant tissues onto Alvetex® scaffold?

It is possible to take explanted tissue and continue to let it grow in 3D in Alvetex® scaffold. The well-insert format is recommended for this purpose.

Is it possible to cryosection Alvetex® scaffold?

This method has not been performed in-house, however customers have reported successful cryosectioning of Alvetex® scaffold.

How long can you culture cells for?

Depending on the cell type and the method of analysis to be performed, Alvetex® scaffold 12 well plates are usually recommended for cell culture experiments lasting between 1 and 10 days. Well inserts are more suited to long term experiments of between 1 and 5 weeks.

Can you perform co-culture experiments?

There are several options available, including;

i) Two or more cell types can be seeded onto one Alvetex® scaffold disc simultaneously;

ii) one cell type can be grown in Alvetex® scaffold and a second cell type can be seeded on top;

iii) cells can be grown in 3D or 2D at the bottom of an Alvetex® scaffold 12 well plate and different cells can be grown above in an Alvetex® scaffold 12 well insert, potentially exerting a paracrine effect upon each other;

iv) two different cell types can be seeded inside separate Alvetex® scaffold discs, then one disc placed on top of the other to bring the two sets of 3D cells into contact with each other.

Is it possible to see the co-culture cells in Alvetex® well inserts growing?

There is not practical way to monitor the 2 cell types during culture unless you want to use some of the well inserts during the optimisation phase. In that case, you can fix, embed and image the 3D cultures using histology or IHC to see what is happening in a visual assay.

Can you perform FACs analysis on cells from Alvetex® scaffold?

Yes - cells removed from Alvetex® scaffold using the partial retrieval protocol can be used for FACs analysis. Please refer to www.amsbio.com/alvetex.aspx or contact info@amsbio.com.
Is Alvetex® scaffold suitable for cell invasion and cell migration assays?

Although formal protocols have not yet been released, it is possible to measure the rate of migration using image analysis of histological sections, or by fluorescently labelling cells.

What happens if more than the recommended number of cells are seeded onto Alvetex® scaffold?

The seeding densities recommended are guidelines and may need to be optimised depending on cell type. If too many cells are seeded, the scaffold may become saturated and cells may migrate out and grow in 2D on the surrounding plastic ware.

What happens if you seed your cells in a larger volume than is recommended?

If larger cell seeding volumes are used then the cells may get drawn through the scaffold with the liquid before they are able to attach, leading to cells adhering to the plastic ware below.

What can I do if my paraffin-embedded section of Alvetex® scaffold does not adhere to the microscope slide?

Poor adhesion can be the result of having limited cell numbers within Alvetex® scaffold, therefore increasing the cell culture incubation time to attain a higher density of cells before processing may help. The use of electrostatic charged slides is also recommended.