REINNERVATE ALVETEX[®] 3D CELL CULTURE SYSTEMS



Achieve genuine three dimensional cell culture, simply and routinely

Alvetex is a highly porous polystyrene scaffold designed for 3D cell culture.

Cells grown in Alvetex maintain their *in vivo* morphology, behaviour and responsiveness within an *in vitro* model system. Alvetex enables cells to maintain their natural shape and to freely interact with neighbouring cells in 3D so that they function in a more physiologically relevant manner.

Presented as a 200 µm thick membrane, Alvetex has been adapted to fit a variety of conventional cell culture plasticware formats. Each product unit has been terminally sterilised by gamma irradiation and remains sterile until its blister pack is opened. Alvetex requires an ethanol wash prior to use to render it hydrophilic. Alvetex does not degrade during normal use.

DIFFERENT ALVETEX ARCHITECTURES TO SUIT YOUR RESEARCH NEEDS

Alvetex is now available in two types: Alvetex Scaffold and Alvetex Strata. Both materials are presented as 200 μm thick membranes of highly porous cross-linked polystyrene. The difference between them is their fine structure and architecture.

Alvetex Scaffold, our market leading product, is primarily designed for three dimensional culture of dissociated mammalian cells within the scaffold. Average void size: 42 µm.

Alvetex Strata, our second generation product, is primarily designed to support the growth of cells and intact tissues on the surface of the membrane. Average void size: 15 μ m.







Scanning electron micrographs of Alvetex Scaffold (left) and Alvetex Strata (right) in transverse section. Scale bars: 100 $\mu m.$

ALVETEX ENHANCES THE BIOLOGICAL RELEVANCE OF YOUR CELL CULTURE RESEARCH

Typical mammalian cells are around $10-25 \ \mu m$ in size and are rarely further than 0-50 μm from another cell or 100-200 μm from a source of nutrients via a blood capillary. By recreating this complex cellular organisation and environment experienced by cells within their native tissues, Alvetex 3D cell culture enables more accurate investigation into the study of cell behaviour and function compared with conventional 2D model systems.

Cells grow and divide occupying the 3D space within Alvetex (or in many instances, on top of Alvetex Strata), maintaining their natural shape and forming complex interactions with one another in a manner that closely mimics normal growth in tissues. The cells may lay down extra-cellular matrix which often leads to the formation of "mini-slab" tissue-like structures. Alvetex is compatible with a broad range of standard molecular, cellular and histological techniques.

Left: Examples of cells grown on Alvetex, visualised by various techniques. (A.) Scanning Electron Micrograph of cells grown throughout Alvetex Scaffold. (B.) Murine keratinocytes grown in Alvetex Scaffold; fixed, embedded, Paraffin sectioned, stained and viewed by bright field microscopy. (C.) Hepatocarcinoma HepG2 cells grown on Alvetex Scaffold. Triple fluorescent stained and viewed by confocal microscopy. (D.) Caco-2 cells in 3D growth of on top of Alvetex Strata.



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MULTIWELL PLATE FORMATS: ALVETEX SCAFFOLD

Alvetex Scaffold 12 Well Plate

Comprised of a single loose disc of Alvetex Scaffold and a polystyrene clip in each well of a 12 well plate. The clip holds the disc in position during transit and use, and can easily be removed for access to the Alvetex Scaffold and cells grown in 3D culture.

The 12 well plate format, a simple presentation of Alvetex Scaffold technology, is primarily suitable for short term culture experiments where the medium is replaced every 1-2 days.





Product	Catalogue code	Presentation
Alvetex [®] Scaffold 12 Well Plate (with lid)	AVP002-2 AVP002-10 AVP002-80	2 × 12 well plates 10 × 12 well plates 80 × 12 well plates

Units are individually sterile blister packed.

Alvetex Scaffold 24 Well Plate

Comprised of a single loose disc of Alvetex Scaffold and a polystyrene clip in each well of a 24 well plate. The clip holds the disc in position during transit and use, and can easily be removed for access to the Alvetex Scaffold and cells grown in 3D culture.

The 24 well plate format, a simple presentation of Alvetex Scaffold technology, is primarily suitable for short term culture experiments where the medium is replaced every 1-2 days.





24 well plate cli

Alvetex disc

Product	Catalogue code	Presentation		
Alvetex [®] Scaffold 24 Well Plate (with lid)	AVP006-2 AVP006-10 AVP006-80	2 × 24 well plates 10 × 24 well plates 80 × 24 well plates		

Units are individually sterile blister packed.



Alvetex Scaffold 96 Well Plate

Comprised of a black 96 well plate, clear plastic base, with Alvetex Scaffold at the bottom of each well. The Alvetex Scaffold has been heat welded to the base of the wells in a process which does not alter its physical structure.

Cells growing in 3D are exposed to culture medium from above only, and therefore predominantly reside in the top portion of scaffold.

Alvetex Scaffold 96 well plate technology is compatible with a wide range of *in vitro* cell viability assays.





Product	Catalogue code	Presentation
Alvetex® Scaffold 96 Well Plate (with lid)	AVP009-2 AVP009-10 AVP009-80	2 × 96 well plates 10 × 96 well plates 80 × 96 well plates

Units are individually sterile blister packed.

Alvetex Scaffold 384 Well Plate

Comprised of a black 384 well plate, clear plastic base, with Alvetex Scaffold at the bottom of each well. The Alvetex Scaffold has been heat welded to the base of the wells in a process which does not alter its physical structure.

Cells growing in 3D are exposed to culture medium from above only, and therefore predominantly reside in the top portion of scaffold.

Alvetex Scaffold 384 well plate technology is compatible with a wide range of *in vitro* cell viability assays.





Product	Catalogue code	Presentation		
Alvetex [®] Scaffold 384 Well Plate (with lid)	AVP010-2 AVP010-10 AVP010-80	2 × 384 well plates 10 × 384 well plates 80 × 384 well plates		

Units are individually sterile blister packed.

WELL INSERT FORMATS: ALVETEX SCAFFOLD AND ALVETEX STRATA

Alvetex 6 Well Insert Format

Comprised of discs of either Alvetex Scaffold or Alvetex Strata in individually sealed polystyrene inserts, designed to fit into most 6 well plates or our custom-made 'Alvetex Well Insert Holder in Deep Petri Dish' (AVP015).

Note that plates and well insert holders are not supplied with the product and have to be sourced separately.

The presentation of Alvetex in well insert formats is versatile, enabling long term 3D culture as cells can receive nutrients from media above and below the membrane, sustaining optimal 3D cell growth.





Product	Catalogue code	Presentation
Alvetex [®] Scaffold 6 well inserts	AVP004-12 AVP004-48 AVP004-96	12 inserts 48 inserts 96 inserts
Alvetex [®] Strata 6 well inserts	STP004-12 STP004-48 STP004-96	12 inserts 48 inserts 96 inserts

Inserts are individually sterile blister packed.

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Alvetex Well Inserts Enable Three Different Media Fill Options

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Media in contact from below only. This enables 3D growth at the air/liquid interface.



Media in contact above and below. Independent compartments enable 3D growth with two different media constituents.



Media in contact above and below. Interconnected compartments enable optimal conditions for maximising cell growth and increased viability.



Alvetex 12 Well Insert Format

Comprised of discs of either Alvetex Scaffold or Alvetex Strata in individually sealed polystyrene inserts, designed to fit into most 6 well or 12 well plates, or our custom-made 'Alvetex Well Insert Holder in Deep Petri Dish' (AVP015).

Snapping the extended wings off Alvetex 12 well inserts (along "break lines") enables fitting into 12 well plates.

Note that plates and well insert holders are not supplied with the product and have to be sourced separately.

The presentation of Alvetex in well insert formats is versatile, enabling long term 3D culture as cells can receive nutrients from media above and below the membrane, sustaining optimal 3D cell growth.





Inserts are individually sterile blister packed.

Alvetex Well Insert Holder and Deep Petri Dish

Comprised of a single well insert holder in a deep Petri dish with lid. The well insert holder is capable of housing up to three Alvetex well inserts (either 6 or 12 well inserts). The Petri dish itself is not tissue culture treated.

The Alvetex Well Insert Holder and Deep Petri Dish enables users to grow their 3D cultures in larger volumes of media compared to an ordinary multiwell plate, facilitating fewer media changes. Capable of sustaining long term 3D culture experiments (3-4 weeks).

The well insert can be positioned at three different levels in the insert holder: high, medium and low. This feature allows cultures to be raised to the air liquid interface by moving the insert to a different level within the same holder.

Positioning the well inserts at different levels may also be used to conserve expensive media or allow for increasing media volumes for demanding cell types over the course of a long term experiment.



Product	Catalogue code	Presentation		
Alvetex [®] Well Insert Holder	AVP015-2	2 units		
and Deep Petri Dish (with lid)	AVP015-10	10 units		

Units are individually sterile blister packed.



Well inserts at different holder levels:



Well insert

on upper level





Well insert on middle level

Well insert on lower level



Alvetex Perfusion Plate

This product allows scientists to create cell based models that are another step closer to the environment experienced by cells and tissue *in vivo*. The systems can also be used to create complex co-cultures, multi-organ systems and to study paracrine effects.

Each unit contains an Alvetex Perfusion Plate with a lid and two Luer locks.





Alvetex Perfusion Plate (with Luer locks)



Example: Alvetex Perfusion Plate showing an Alvetex 6 Well Insert in position

Product	Catalogue code	Presentation		
Alvetex [®] Perfusion Plate	AVP011-2	2 units		
(with lid)	AVP011-10	10 units		

Units are individually sterile blister packed. **Note:** Pump and tubing is not included.



Example: Alvetex Perfusion Plate showing 4 × Alvetex 12 Well Inserts in position



Example: Alvetex Perfusion Plate showing an Alvetex 12 Well Insert in position

Alvetex Kits

Product	Catalogue code	Presentation
Alvetex [®] Scaffold Plate Starter Kit	AVP-KIT-1	1 × 12 well plate 1 × 24 well plate 1 × 96 well plate
Alvetex [®] Scaffold Well Insert Starter Kit	AVP-KIT-2	6 × 6 well inserts 6 × 12 well inserts 1 × Alvetex Well Insert Holder in a deep Petri dish
Alvetex [®] Strata Well Insert Starter Kit	STP-KIT-2	6 × 6 well inserts 6 × 12 well inserts 1 × Alvetex Well Insert Holder in a deep Petri dish

Product	Catalogue code	Presentation
Kit: Alvetex [®] Perfusion Plate with Alvetex [®] Scaffold 6 well inserts	AVP-KIT-3	2 × Alvetex Perfusion Plates with Luer locks 12 × Alvetex Scaffold 6 well inserts
Kit: Alvetex [®] Perfusion Plate with Alvetex [®] Scaffold 12 well inserts	AVP-KIT-4	2 × Alvetex Perfusion Plates with Luer locks 12 × Alvetex Scaffold 12 well inserts
Kit: Alvetex [®] Perfusion Plate with Alvetex [®] Scaffold 6 well inserts	AVP-KIT-5	5 × Alvetex Perfusion Plates with Luer locks 48 × Alvetex Scaffold 6 well inserts
Kit: Alvetex [®] Perfusion Plate with Alvetex [®] Scaffold 12 well inserts	AVP-KIT-6	5 × Alvetex Perfusion Plates with Luer locks 48 × Alvetex Scaffold 12 well inserts

Choosing the right Alvetex format based on assay type

The table below can guide your choice of the most suitable Alvetex format for your assay.

	Alvetex Scaffold				Alvetex Strata			
Types of assay	6 well inserts	12 well inserts	12 well plates	24 well plates	96 well plates	384 well plates	6 well inserts	12 well inserts
Viability/Proliferation/ Metabolic Activity Assays	+++	+++	+++	+++	+++	+++	+++	+++
Toxicity Assays	+++	+++	+++	+++	+++	+++	+++	+++
Gene Expression assays (qPCR/microarray)	+++	+++	+++	+++	+++	+++	+++	+++
Protein Expression assays (e.g. western blot)	+++	+++	+++	+++	+++	+++	+++	+++
Air-liquid Interface assays	+++	+++	N/A	N/A	N/A	N/A	+++	+++
Cell Signalling assays	+++	+++	+++	+++	+++	+++	+++	+++
Permeability assays	+++	+++	N/A	N/A	N/A	N/A	+++	+++
Transfection assays	+++	+++	+	+	+	+	+++	+++
Co-culture assays	+++	+++	++	++	++	++	+++§	+++ [§]
Invasion assays	+++	+++	+	+	+	+	++§	++ [§]
Migration assays	+++	+++	+	+	+	+	++§	++ [§]
Histology	+++	+++	++	++	++	+	+++	+++
Immunostaining (IHC/IF)	+++	+++	++	++	++	+	+++	+++
Confocal microscopy	+++	+++	++	++	++	+	++	++
Live cell imaging*	+++	+++	++	++	++	+	++	++
Ex vivo tissue maintenance	+++	+++	++	++	+	+	+++	+++
Live cell retrieval**	++	++	++	++	++	++	++	++

Suggested guidelines for the use of Alvetex formats for cell applications and assays.

- +++ = most suitable
- ++ = suitable
 - = least suitable
- N/A = not applicable

Ranking is based on Alvetex Scaffold disc format suitability, the likely cell yields and therefore signal generation, and whether exogenously added chemicals/ cells can be contained to only one side of the membrane.

*The growth of cells cannot be followed by traditional light microscopy as in 2D, but as with *ex vivo* tissues, 3D structures have to be evaluated using histology or confocal microscopy. Alternatively cell proliferation can be monitored using a viability assay such as the MTT.

**The exact number of cells retrieved from Alvetex varies with the invasiveness of the cell line cultured, e.g. epithelial vs. fibroblastic. Although the three-dimensional structure of Alvetex precludes all 100% of the cells from being routinely retrieved, calls can be retrieved in adequate numbers for quantitative downstream processes, e.g. flow cytometry.

[§] When designing co-culture, invasion or migration set-ups for Alvetex Strata, please keep in mind that some cell lines (e.g. epithelial) have a tendency to multilayer on top of the substrate rather then invade into it.

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AMERICAS • HUMAN TISSUE 9000 Virginia Manor Road, Suite 207 Beltsville, MD 20705, USA Tel: (301) 470-3362 Email: info@bioserve.com

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`***₩***stemgent

AMERICAS • REAGENTS 4 Hartwell Place Lexington, MA 02421, USA Tel: (617) 245-0000 Email: info@stemgent.com

REPROCELL

ASIA PACIFIC KDX Shin-yokohama381 Bldg9F 3-8-11, Shin-yokohama, Kohoku-ku, Yokohama, Kanagawa 222-0033, Japan Tei, +81 45 475 3887 Email: Info_jp@reprocell.com www.reprocell.com

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EUROPE NETPark Incubator Thomas Wright Way Sedgefield, Co. Durham TS21 3FD, UNITED KINGDOM Tel: +44 (0)1740 625 266 Email: info@reinnervate.com

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AMERICAS & EUROPE HUMAN TISSUE ASSAYS Weipers Centre, Garscube Estate Bearsden Road, Glasgow G61 1QH United Kingdom Tel: +44 (0)141 330 3831 Email: info@biopta.com www.biopta.com

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