

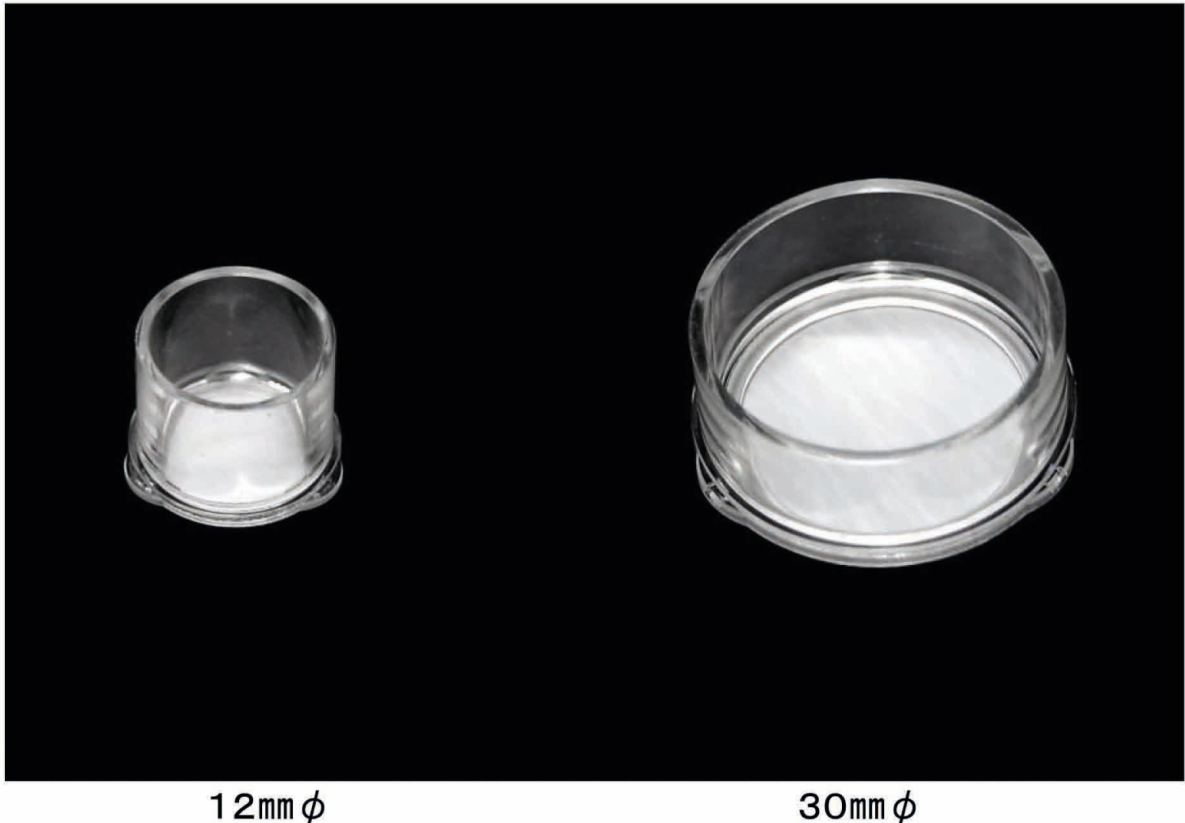


***From now on, First Choice for Cell***

***Culture is VECCELL-3D Insert !!***

A New 3 Dimensional Cell Culture Insert which is so unique and versatile is commercially available in 2007. You can see easily the difference between conventional plastic dish cell culture and our new product – Vecell cell culture.

### **VECELL-3D Insert**



12mm  $\phi$

30mm  $\phi$

## **Natural cell culture almost like in vivo on VECCELL-3D Insert**

There is a need for a surface upon which cells behave as though in vivo, thus the surface provides a relevant in vitro model of cellular processes. This has clear advantages in replacing animal models of disease, target validation, and toxicology.

**Vessel Inc.** thinks cell culture using permeable micro-porous membranes will become a standard method for culturing cells. The membranes bring about a great improvement in culturing polarized cells since these permeable supports permit cells to uptake and secrete molecules on both their basal and apical surfaces and thereby carry out metabolic activities in a more natural fashion.

**Vessel Inc.** declares that the porosity of membrane is so important for cell function and proliferation, but none of currently available scaffolds has enough porous structures.

**Vessel Inc.** also declares that many cell-based assays in use today which rely on two-dimensional (2D) flat substrates can not emulate in vivo three-dimensional (3D) physiological conditions.

Dr.Kodama(Managing Director and Founder ) has developed a technology which enables modification of highly hydrophobic surfaces to hydrophilic ones. Using the technology **Vessel Inc.** has developed a unique highly permeable micro-porous membranes of fibrous configuration whose surface is modified with ECM(collagen) and the technology leads to a New 3 Dimensional Cell Culture Insert- **VECELL-3D Insert**. **VECELL-3D Insert** allows cell adhesion and proliferation in a physiological conditions without any stress unlike surface of conventional plastic cell culture surfaces as shown in Fig.1. Cells on Vecell can adhere on membrane in a 3D fashion as shown in Fig.2.

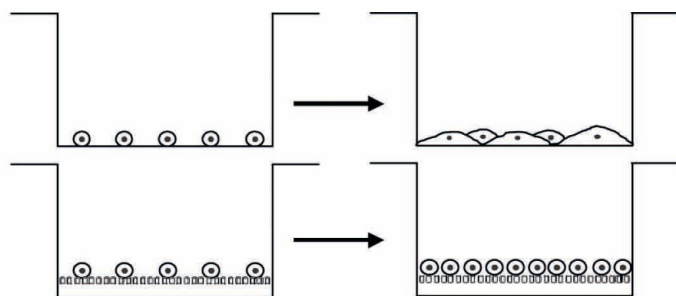


Fig.1  
Difference between normal plastic dish culture and VECCELL membrane culture  
Upper: Cell culture on normal plastic dish  
Below: Cell culture on Vecell

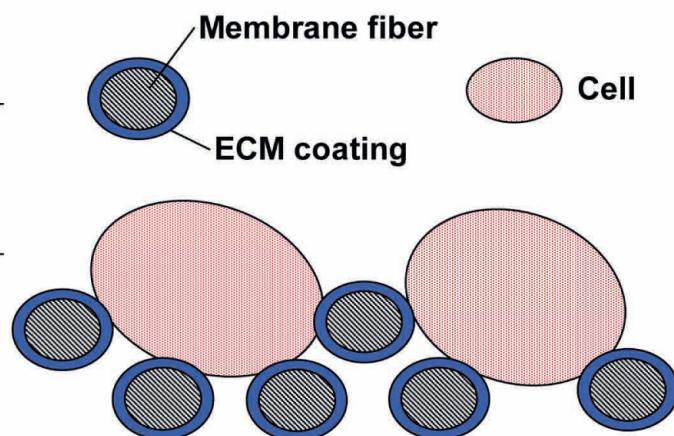


Fig. 2  
Schematic Model for Cell Culture on VECCELL Membrane



## Features of Vecell

- ①The membrane of **VECELL-3D Insert** is homogenously thin-layer covered with cell adhesive materials (Figs. 3 and 4). So you can directly seed cells on the membrane without further modification.
- ②The membrane structure is so porous that the medium is easily passed through the membrane to mimic in vivo environment . So **VECELL** can be used for permeability assay (Fig.4).
- ③Cell proliferation on **VECELL** is in a “natural state” without any stress. In contrast cells on a surface of conventional plastic ware spread due to the strong interaction between 2D-solid plastic surface and cells(Fig.1). so cell nuclei are observed due to unnatural cell flatness(Fig.5).
- ④The cells cultured on **VECELL** can be visualized by a phase contrast microscope (Fig.5). For viewing live cells, microscopic observations can be made through the bottom of multiwell plate containing **VECELL** and media.

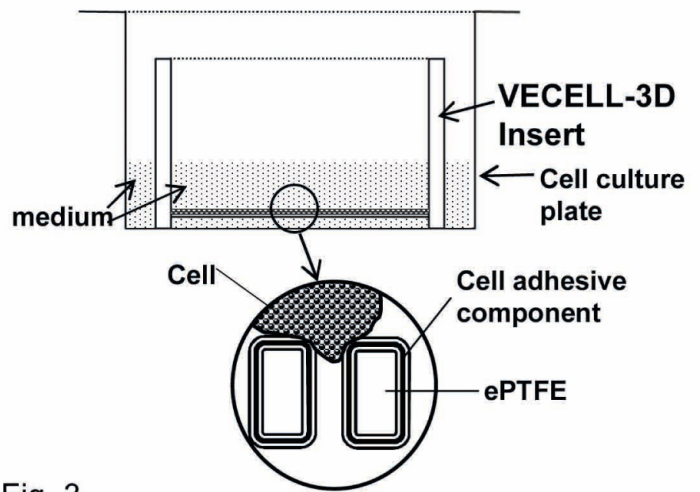


Fig. 3  
The membrane of **VECELL** is already coated with ECM (collagen).

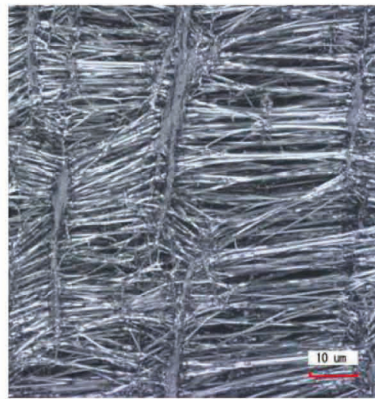


Fig. 4  
A photograph of **VECELL** membrane under SEM.

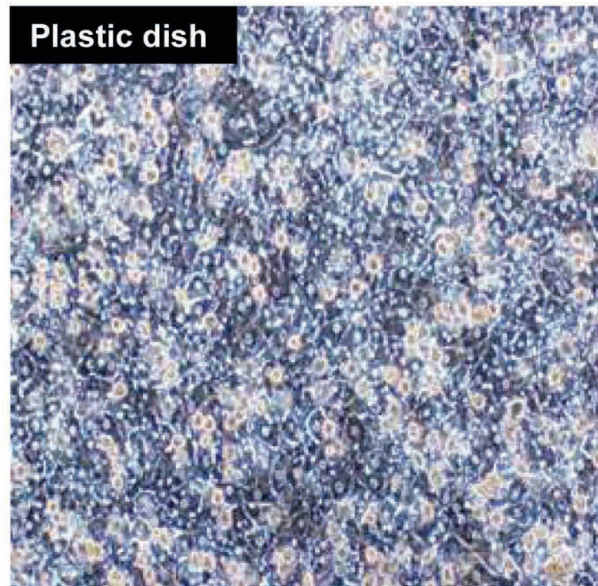
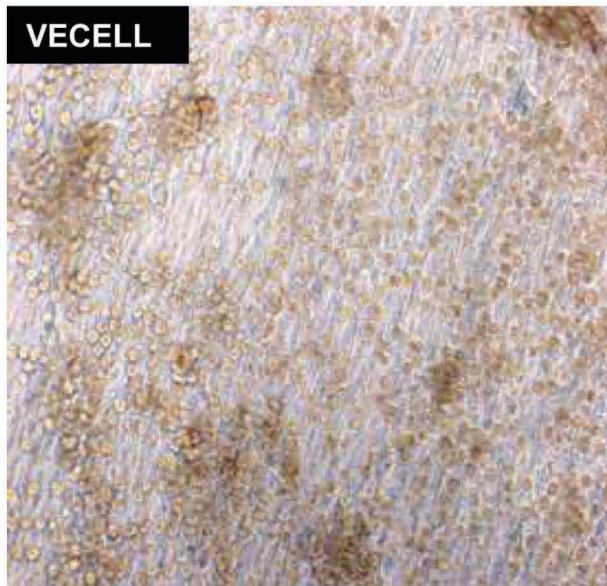


Fig. 5  
The phase contrast light-microscope pictures of Rat Hepatocytes. The results of 3 days Culture. Left : on **VECELL** Right :on conventional plastic dish



# Experimental example

## ① Long period stability of Cell Culture on Vecell

Cell growth of L929 cells on **VECELL** was compared with that on a conventional plastic dish (Fig.6). L929 cells on a conventional plastic dish proliferate to confluency and then detach from the surface. In contrast cells on **VECELL** are viable even after 264 hrs culture period(Fig.7).

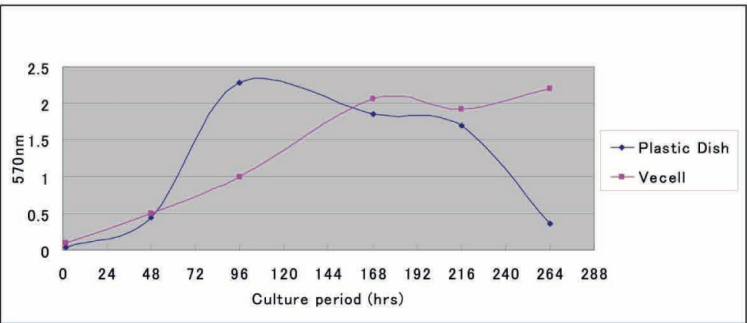
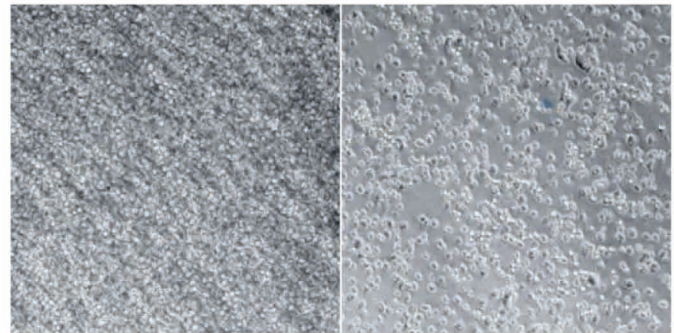


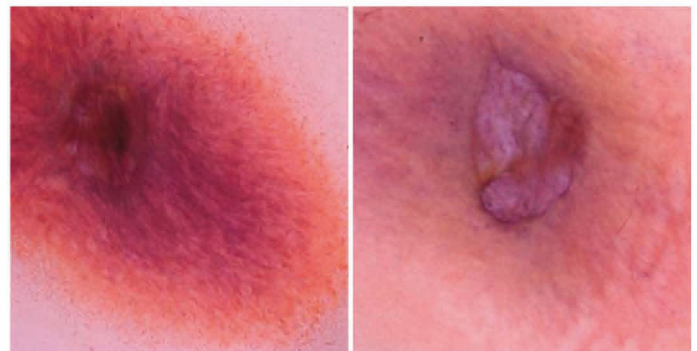
Fig. 6  
MTT assays for cell proliferation on **VECELL** and conventional plastic dish



**VECELL** **Plastic Dish**  
Fig. 7  
Phase Contrast Micrographs after 264 hrs Cell culture

## ② Application for Tissue Culture

Human bone membrane tissue was placed on **VECELL** and on a conventional plastic dish. Actively differentiated cells are shown in Fig.8. ALP ( alkaline phosphatase) expression stained with purple color are clearly observed on **VECELL** membrane. This result indicates that **VECELL** can be applied for tissue culture.



**VECELL** **Plastic Dish**  
Fig. 8  
The result of Human Bone Membrane Tissue culture on **VECELL** or on a plastic dish. ALP was stained and tissue was observed by a stereoscopic microscope (Courtesy of Dr. T. Kawase of Niigata University).



## 【 VECCELL-3D Insert】

	Diameter	Filter Material	Fibril Length	ECM
① Vecell 30 mm $\phi$ (For 6 Well Cell Culture Plate) [ 12 Quantity / Package ]	30 mm	Poly-tetrafluoroethylene	20-80 $\mu$ m	ocean collagen
② Vecell 12 mm $\phi$ (For 24 Well Cell Culture Plate) [ 24 Quantity / Package ]	12 mm	Poly-tetrafluoroethylene	20-80 $\mu$ m	ocean collagen

**VECELL** is available for 24- and 6-well plate. It is easily prepared for SEM and TEM, and is compatible with cellular and fluorescent stains. All inserts are individually wrapped and sterilized with EO gas.

### General Direction for Use

- ① **VECELL-3D Insert** is already sterilized with EO gas. Use it before 1 year after the manufacture. Lot number denotes the date of manufacture. For example, Lot# 080926 means that the date of manufacture is September 26<sup>th</sup>, 2008. So the expired date is September 25<sup>th</sup>, 2009.
- ② Using a sterile pipette syringe, add the appropriate volume of culture medium to allow the whole surface area of membrane get wet and transparent. And then pipette an appropriate number of cells dispersed in medium and seed cells on to Vecell membrane.
- ③ Remove bubbles if you find out them below the membrane.
- ④ Cultured cells on **VECELL** membrane can be observed by a phase contrast microscope.