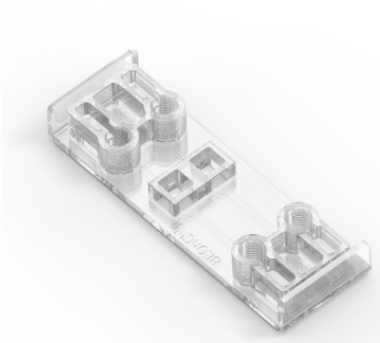
BE-TRANSFLOW
TECHNICAL CHARACTERISTICS

BEOnChip
Biomimetic Environment On Chip



BE-TRANSFLOW is a slide format versatile microfluidic device for cell culture under biomimetic conditions. It allows a combination of a 2D-3D organized culture with the possibility of establishing flow and Air Liquid Interface (ALI). Our most biomimetic microdevice to copy *in vitro* different tissue structures.

Examples of applications are immune system *in vitro* model, cancer-metastasis *in vitro* model, skin and gut on chip model.

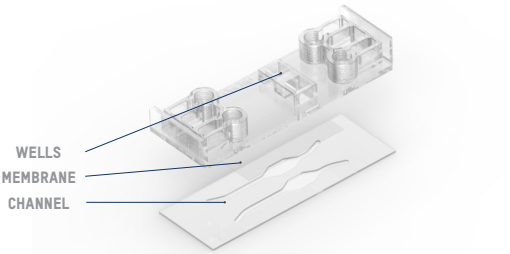
For further information, please contact BEONCHIP

MATERIAL

BE-TRANSFLOW chips are made of biocompatible plastic and are gas-impermeable, for effective gradients of CO_2, O_2, etc. They have excellent optical properties, with high transparency and low auto-fluorescence.

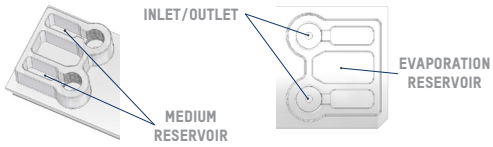
TECHNICAL FEATURES

The design of the transflow consists of 2 independent wells with a porous membrane as a bottom. This membrane is connected with a lower channel. Flow is possible in the channel with the help of screw-like designed inlet and outlet wells by screwing them with connectors and tubes to a fluidic system. Evaporation reservoirs are next to the medium reservoirs to be filled with PBS or water during the incubation before closing the system with tubes.



	Height	Width	Lenght	Total Volume
Channel	375 µm	1'5 mm	45 mm	43,5 µL
Well	6 mm	5'7 mm	5'7 mm	195 µL
Inlet/Outlet	7 mm	UNF 1/4"- 28		130 µL
M. Reservoir	5 mm	3'6 mm	8'8 mm	185 µL
Membrane	Pore size: 1µm			

For personalized membranes, contact BEONCHIP



CONTENT

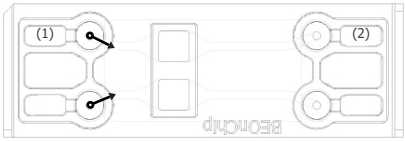
The product reaches the user sterilized (10 Be-Transflow per box). It can be stored in dry places which are not exposed to direct sunlight at room temperature (15-25ºC).

CELL CULTURE COATING

BE-TRANSFLOW chips have been treated to obtain a hydrophilic surface that facilitates filling the devices with aqueous solutions and/or gels and promotes cell adhesion.

In case of a certain coating is required, prepare your coating solution (Collagen I, Collagen IV, Fibronectin, Poly-L-Lysine, Poly-D-Lysine…) according to the manufacturer's instructions and apply it into the channel. Aspirate the channel and wash with distilled water or PBS to remove excess coating solution by using 5-10 times the volume of the channel.

FILLING AND HANDLING



1. Trypsinize and count cells as usual. Cell concentration will vary with the cell type. It is recommended to seed a cell concentration to obtain a confluent layer within 2-3 days.

2. Seed cells. It is possible to seed a monolayer, a hydrogel or both.

2A. In the upper channel over the membrane.

2B. It is even possible to culture a monolayer below the membrane filling the lower channel with cells and turning it over to allow them to attach. If you want to perform this kind of assay, seed the cells first in the lower channel and, when the cells are attached, flip the device and proceed with the protocol to seed the cells in the upper one.

To do this, with a P-100 or P-1000 pipette, take 100 µL of medium with cells and add it in the inlet. Leave the medium with cells flow through the channel letting it reach the outlet. Pay attention to fill the inlet (1) and leave the outlet (2) empty to allow the pressure difference equalises the medium among inlet and outlet. It is important to add water or PBS in the evaporation reservoirs to avoid evaporation.

3. Let cells adhere to the surface (the time may vary depending on the cell type).

4. If you are not mounting the flow system on the same day of seeding, add the desired amount of medium in the medium reservoirs. Depending on your cell type, pay attention to change the medium. To do this, medium is added carefully in the medium reservoir next to the inlet and the pressure difference will equalise the medium among inlet and outlet.

5. In order to connect the device to a flow system, screw the connectors into the outlet and inlet (in that order). Both inlets and outlets are designed to be screwed to connectors (1/4" - 28). Remember that the device and the tubes have to be primed with medium before connecting.

It is possible to use a rocker to move medium from one reservoir to another. With a final volume of 100 µl per reservoir it is possible to apply 45° of inclination.

ASSEMBLY OF THE FLOW SYSTEM

Beonchip has some advices to set the flow system configuration.
Previous considerations:

1. Prewarm the tubes and the fluidic elements during 15-20 minutes at the incubator.
2. Set the system in a laminar flow cabinet.
3. Cells must be well adhered to the surface before mounting the flow system.
4. The device should be never left without culture medium inside or in the inlets/outlets.

To assemble the flow system, the following will be considered:

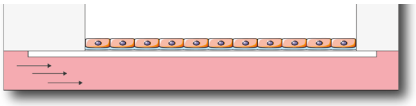
1. Fill the inlet well completely with medium so that no air bubbles remain.
2. Prime the system of tubes that connect to the inlet before assembling the system.
3. Both inlets and outlets are designed to be able to use connectors (1/4" - 28). Once this tube system is primed, the connector can be screwed into the inlet. At this point, extreme care must be taken to ensure that no air bubbles enter the system.
4. Finally, the tube is connected to the outlet and thus the system is closed.
5. Check that there are no leaks in the system. To do this, leave the pump running for a couple of minutes before placing the devices in the bioreactor or in the incubator.

CELLS SEEDING

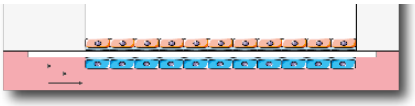
Below there are some examples of use for a better understanding of the possibilities of the device.

Depending on the assay, either monoculture or coculture can be done. Seeding a monolayer over and/or below de membrane (1 and 2) with different cell types. For more realistic assays it is possible to add an hydrogel cell culture above the monolayer (3).

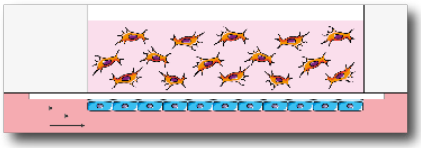
(1)



(2)



(3)



PREPARATION FOR CELL MICROSCOPY

It is possible to monitor fixed or living cells and also chemical gradients. Most of the monitoring systems used in traditional cell culture can be taken to BEONCHIP microfluidic devices. Common fixatives can be used. Cell viability can be evaluated using different dyes. Moreover, immunofluorescent staining can be performed to identify specific targets. Also, cell cycle fluorescent reporters can be used.

Please contact BEONCHIP for further assistance.

OTHER READOUTS

Recovering the cells is possible in order to perform downstream applications such as flow cytometry, RNA extraction (PCR)...

Please contact BEONCHIP for further assistance.

To prevent bubbles from forming during filling, please firstly prewarm the device and avoid empty completely tips of pipettes. Hold the plunger firmly while removing the pipette from the inlets so that the negative pressure will not suck the solutions up.