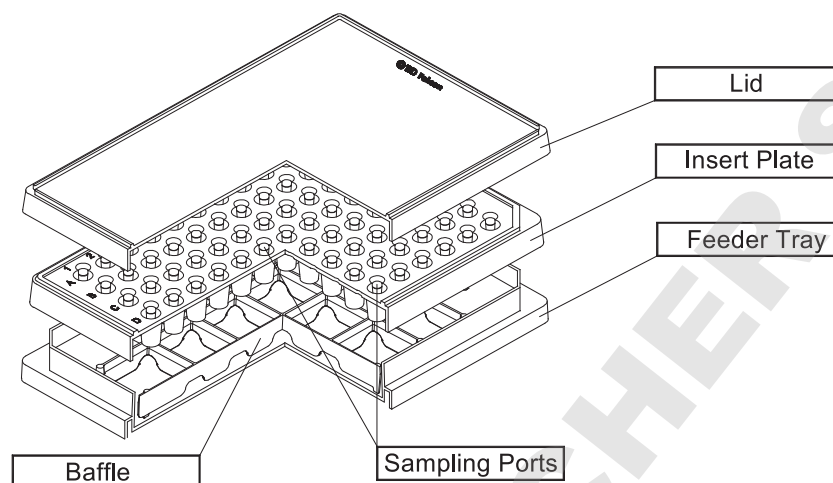


## Falcon® 96-Multiwell Insert System



### Product Use Guidelines for Manual or Automated High Throughput Screening Assays

The Falcon 96-Multiwell Insert System is a cell culture insert platform suitable for both manual and robotic screening of compounds in cell-based assays. The system has been tested for its ability to produce a differentiated monolayer of Caco-2, LLC-PK1, and MDCK cells making it an ideal platform for *in vitro* bioavailability and permeability studies.

This automation compatible platform is composed of a 1.0  $\mu\text{m}$  pore size PET (polyethylene terephthalate) membrane-based 96-Multiwell Insert Plate, a media Feeder Tray and Baffle or 96-Square Well, Angled-Bottom Plate and Lid. All 96 wells are integrated into a one-piece plate, which fits into a Feeder Tray, suitable for making media changes in one transfer. To analyze individual samples, simply transfer the Insert Plate into the Falcon 96-Square Well, Angled-Bottom Plate. If desired, the Falcon 96-Square Well, Angled-Bottom Plate may also be used for culturing the cells, thus eliminating the transfer step from the single-well Feeder Tray for individual sample analysis.

### Product Specifications: Falcon 96-Multiwell Insert Systems

Corning Cat. No.	Description	Optical Quality	Included with each Insert Plate	Plates per pack
351130	Falcon 96-Multiwell Insert System	Transparent	Feeder Tray, Baffle with Lid	1
351131	Falcon 96-Multiwell Insert System	Transparent	Feeder Trays, Baffles with Lids	5
353938	Falcon 96-Multiwell Insert System	Transparent	96-Square Well, Angled-Bottom Plates with Lids	5
353924	Additional Falcon Feeder Trays, Baffles with Lids			5
353925	Falcon 96-Square Well, Angled-Bottom Plates with Lids			5

All membranes are track-etched PET (Polyethylene Terephthalate). All products are sterilized by gamma irradiation and are intended for single use only.

\* Feeder Tray and 96-Square Well, Angled-Bottom Plates are made of Polystyrene and have non-treated surfaces.

Falcon® 96-Multiwell Insert System: Plate Dimensions

**LID:** Cat. Nos.: 351130, 351131

Material: PS (Polystyrene)  
Length: A = 128.1 mm (5.045 inches)  
Width: B = 85.5 mm (3.367 inches)  
Height: C = 7.4 mm (.290 inches)

**INSERT HOUSING:**

Material: PET (Polyethylene Terephthalate)  
Length: D = 128.1 mm (5.045 inches)  
Width: E = 85.5 mm (3.367 inches)  
Height: F = 7.4 mm (.290 inches)

**Insert Wells:**  
Top Interior Diameter: 5.7 mm (.226 inches)  
Bottom Interior Membrane Diameter: 3.2 mm (.125 inches)  
Bottom Exterior Diameter: 5.2 mm (.206 inches)  
Total Well Depth: 10.4 mm (.410 inches)  
Membrane Distance from Tray: 3.0 mm (.120 inches)  
Well to Well Distance: 9.0 mm (.354 inches)  
Sampling Port Diameter: 2.8 mm (.110 inches)

**BAFFLE:**

Material: PS (Polystyrene)  
Length: G = 118.2 mm (4.654 inches)  
Width: H = 75.8 mm (2.986 inches)  
Height: J = 8.9 mm (.350 inches)

**FEEDER TRAY:**

Material: PS (Polystyrene)  
Length: K = 127.8 mm (5.030 inches)  
Width: L = 85.5 mm (3.365 inches)  
Height: M = 14.4 mm (.565 inches)  
Flange Height: N = 6.4 mm (.250 inches)

**LID:** Cat. No.: 353924

Material: PS (Polystyrene)  
Length: A = 128.1 mm (5.045 inches)  
Width: B = 85.5 mm (3.367 inches)  
Height: C = 7.4 mm (.290 inches)

**BAFFLE:**

Material: PS (Polystyrene)  
Length: G = 118.2 mm (4.654 inches)  
Width: H = 75.8 mm (2.986 inches)  
Height: J = 8.9 mm (.350 inches)

**FEEDER TRAY:**

Material: PS (Polystyrene)  
Length: K = 127.8 mm (5.030 inches)  
Width: L = 85.5 mm (3.365 inches)  
Height: M = 14.4 mm (.565 inches)  
Flange Height: N = 6.4 mm (.250 inches)

**LID:** Cat. No.: 353938

Material: PS (Polystyrene)  
Length: A = 128.1 mm (5.045 inches)  
Width: B = 85.5 mm (3.367 inches)  
Height: C = 7.4 mm (.290 inches)

**INSERT HOUSING:**

Material: PET (Polyethylene Terephthalate)  
Length: D = 128.1 mm (5.045 inches)  
Width: E = 85.5 mm (3.367 inches)  
Height: F = 7.4 mm (.290 inches)

**Insert Wells:**  
Top Interior Diameter: 5.7 mm (.226 inches)  
Bottom Interior Membrane Diameter: 3.2 mm (.125 inches)  
Bottom Exterior Diameter: 5.2 mm (.206 inches)  
Total Well Depth: 10.4 mm (.410 inches)  
Membrane Distance from Tray: 3.0 mm (.120 inches)  
Well to Well Distance: 9.0 mm (.354 inches)  
Sampling Port Diameter: 2.8 mm (.110 inches)

**ANGLED-BOTTOM PLATE:**

Material: PS (Polystyrene)  
Length: P = 127.8 mm (5.030 inches)  
Width: Q = 85.5 mm (3.365 inches)  
Height: R = 15.6 mm (.613 inches)  
Flange Height: S = 6.4 mm (.250 inches)

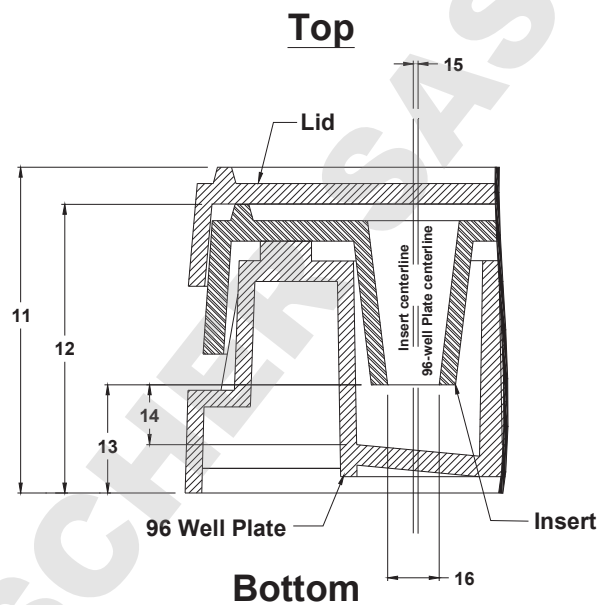
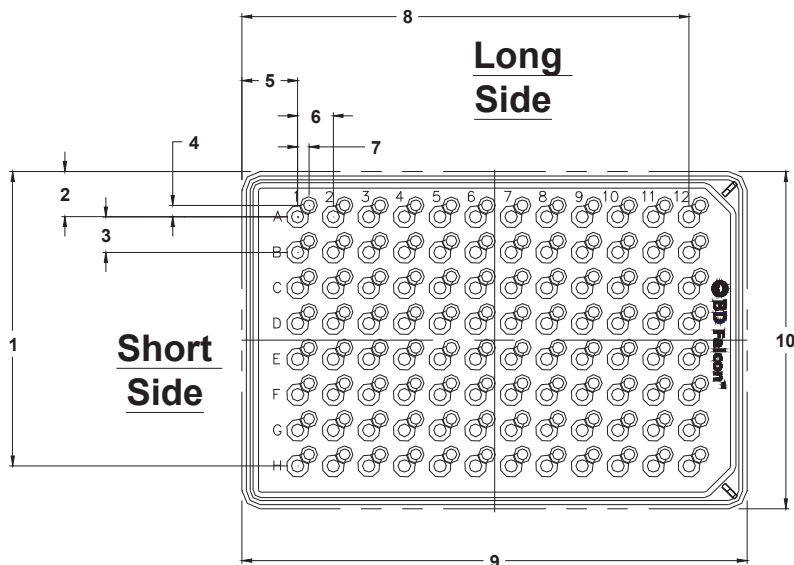
**LID:** Cat. No.: 353925

Material: PS (Polystyrene)  
Length: A = 128.1 mm (5.045 inches)  
Width: B = 85.5 mm (3.367 inches)  
Height: C = 7.4 mm (.290 inches)

**ANGLED-BOTTOM PLATE:**

Material: PS (Polystyrene)  
Length: P = 127.8 mm (5.030 inches)  
Width: Q = 85.5 mm (3.365 inches)  
Height: R = 15.6 mm (.613 inches)  
Flange Height: S = 6.4 mm (.250 inches)

Suggested Media Volume in Insert	50-75 µl
Suggested Media Volume in 96-Square Well, Angled-Bottom Plate (basolateral)	260 µl
Suggested Media in Feeder Tray	25-30 ml
Suggested Media when using/culturing cells in a 96-Square Well, Angled-Bottom Plate	260 µl
Effective Diameter of Membrane	3.2 mm
Effective Growth Area of Membrane	0.0804 cm <sup>2</sup>
Effective Growth Area in 96-Square Well, Angled-Bottom Plate (per well)	0.64 cm <sup>2</sup>
Pore Density: 1.0 µm	1.6 x 10 <sup>6</sup> pores/cm <sup>2</sup>
Insert Height (apical)	10.4 mm



**Falcon® 96-Multiwell Insert System: Plate Indexing**

1	Long Side of 96-Square Well, Angled-Bottom Plate to well Center H1 of 96-Multiwell Insert Plate	74.51 mm
2	Long Side of 96-Square Well, Angled-Bottom Plate to well Center A1 of 96-Multiwell Insert Plate	11.52 mm
3	Well-to-Well Centers (Center of one well to the center of another)	9.00 mm
4	Well Center of Insert Membrane to Center of Insert Sample Port	2.89 mm
5	Short Side of 96-Square Well, Angled-Bottom Plate to well Center A1 of 96-Multiwell Insert Plate	14.10 mm
6	Well-to-Well Centers (Center of one well to the center of another)	9.00 mm
7	Well Center of Insert Membrane to Center of Insert Sample Port	2.89 mm
8	Short Side 96-Square Well, Angled-Bottom Plate to well Center A12 of 96-Multiwell Insert Plate	113.08 mm
9	Overall Length of 96-Square Well, Angled-Bottom Plate	127.76 mm
10	Overall Width of 96-Square Well, Angled-Bottom Plate	85.47 mm
11	Total Assembly Height (96-Multiwell Insert Plate, 96-Square Well, Angled-Bottom Plate w/ Lid) Total Assembly Height (96-Multiwell Insert Plate, Feeder Tray w/ Lid)	20.27 mm 19.05 mm
12	Assembly Height (96-Multiwell Insert Plate, 96-Square Well, Angled-Bottom Plate w/o Lid) Assembly Height (96-Multiwell Insert Plate, Feeder Tray w/o Lid)	17.98 mm 16.76 mm
13	Distance from Membrane to Bottom of 96-Square Well, Angled-Bottom Plate Distance from Membrane to Bottom of Feeder Tray	6.55 mm 5.34 mm
14	Distance from Membrane to Bottom of Well of 96-Square Well, Angled-Bottom Plate (High Side Inner) Distance from Membrane to Bottom of Well of 96-Square Well, Angled-Bottom Plate (Low Side Inner) Distance from Membrane to Bottom of Feeder Tray (Inner)	2.87 mm 4.27 mm 3.05 mm
15	Well Center of Insert Membrane to Well Center of 96-Square Well, Angled-Bottom Plate	0.28 mm
16	Usable Membrane Diameter	3.2 mm

## Guidelines for Automated Use

Handle all inserts under aseptic conditions.

If you plan to use this product with a robot fluid handling workstation, please note the following:

1. The sides of the Falcon<sup>®</sup> 96-Multiwell Insert System have been designed so that robotic arms can lift the entire assembly. The lid can be removed by either robotic grippers or by suction from the top. We recommend careful placement of the lid onto the insert plate to prevent knocking the Feeder Tray, which may result in splashing the media.
2. When moving the Falcon 96-Multiwell Insert System using a robotic arm, make sure that the movements are slow and smooth. In most cases, intermediate positions can be programmed to help minimize long movements that are followed by sudden stops. Sudden stops may result in excessive media splashing. Most robotic grippers can be programmed to move at various speeds for XYZ and rotational movement. It is advised that rotational movements should be slower than what is typically used for standard microplates.
3. The Falcon 96-Multiwell Insert System is designed so that media can be added or removed through the access ports without removing the insert plate. If using a 4 or 8 tip probe, it is best to stagger the well positions, rather than adding or removing from a solitary port location. When removing media from the Feeder Tray, it is best to aspirate a greater total volume than what was dispensed in order to minimize residual media. On some media exchange systems it is necessary to first remove the insert plate before exchanging media.
4. Depending on the gripper finger features, it may be necessary to first de-lid the plate when lifting the insert plate out of the Feeder Tray. Some fingers have screws or “teeth” that can be extended so that the lid does not need to be removed while manipulating the Insert Plate. The Insert Plate is designed to be placed on the Falcon 96-Square Well, Angled-Bottom Plate (Cat. No. 353925) in one unique orientation to prevent cross contamination of wells. To properly align the Insert Plate in the 96-Square Well, Angled-Bottom Plate, make sure the Falcon logo on the top of both pieces face the same direction.
5. When used with the Falcon 96-Square Well, Angled-Bottom Plate, sampling of the basalateral (beneath the membrane) may be done with most “fixed” and disposable 96-well pipet tips. Some 200 µl disposable pipet tips with large diameters cannot reach the bottom of the basalateral chamber; however, most of the instrument vendors supply smaller volume tips that are fully compatible. When sampling the apical side (top of the membrane), tips may be indexed between wells using the traditional 9 mm pitch. When sampling from the apical side, make sure the tips do not puncture the membranes. It is best to first tune the XY pipet positions above the Insert Plate, and then adjust the aspirate and dispensing heights using small increments.
6. In order to define both the apical and basalateral pipetting locations, it may be necessary to edit the deck or carrier position file to accommodate multiple plates in the same location. Some instruments require you to overlay individual plate definition files. If this is the case, it is best to define the access ports and the feeder ports separately. Height offsets may be required to properly manipulate or de-lid the Insert Plate at these locations. Also, it may be necessary to toggle between deck layouts in the same protocol. Due to the location of the bottom of the apical well and the bottom of the basalateral well, it may be necessary to turn off associated features that perform liquid volume tracking based on well geometry.

For additional information regarding defining robotic protocols or for a list of compatible pipet tips, please contact Corning Life Sciences Technical Service at 1-800-492-1110; 1-978-442-2200; email [CLSTechServ@corning.com](mailto:CLSTechServ@corning.com).



## General Hints for Using Membrane Insert Products

Handle all inserts under aseptic conditions. Use pre-warmed media to maintain physiological stasis.

**Seeding of Inserts:** Fill the Insert Plates with cells and media before filling the bottom wells with media. This prevents bubbles from getting trapped underneath the insert and prevents cell monolayers or coatings on the membrane from being disturbed by hydrostatic pressure from media below the insert. To determine the optimal seeding density for your cell type on a porous growth surface, we recommend using a range of seeding densities (cells/cm<sup>2</sup>) that brackets the seeding density used on nonporous surfaces (Flasks, Dishes and Plates). For example, if you currently seed at 1.0x10<sup>5</sup> cells/cm<sup>2</sup>, seed at 0.5x10<sup>5</sup>, 1.0x10<sup>5</sup> and 5.0x10<sup>5</sup> cells/cm<sup>2</sup> to determine the optimal initial seeding density.

**Culturing Cells:** Add pre-warmed culture medium to the Feeder Tray. For best results, we recommend adding 25 ml media to the tray. Too little media will prevent contact with the insert. Excess media may slosh out of the Feeder Tray during handling. The Falcon® 96-Square Well, Angled-Bottom Plate may be used in place of the Feeder Tray. For best results, limit media volume in the lower wells to 260 µl. Excess media (>260 µl) will overflow into the insert.

**Feeding (using Feeder Tray):** If desired, any of the 96 top sample ports may be used for aspirating off old media and adding fresh media. The 96 top sample ports will accept an aspirating pipet tip. If used manually, the Insert Plate may be lifted under aseptic conditions for direct access to the Feeder Tray. Replacement Feeder Trays are available separately for added user convenience (Cat. No. 353924).

**Sample Collection (using the 96-Square Well, Angled-Bottom Plate):** The Falcon 96-Square Well, Angled-Bottom Plate (sold separately, Cat. No. 353925) has been specifically designed for use with the Falcon 96-Multiwell Insert System. Each well of the Falcon 96-Square Well, Angled-Bottom Plates features an angled bottom that allows for complete aspiration or sample collection from the bottom of the well without the need to tilt the plate. **Note:** *Standard 96-well plates are not compatible with the Falcon 96-Multiwell Insert System. Use of standard 96-well plates will result in media wicking up on the sides of the wells and possibly into the insert or out of the well.*

**Handling of the Insert:** Once the Insert Plate is placed into a receiver plate containing solution, **do not tilt the system**, this can result in wicking of the basolateral solution.

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