User Guide

Millicell®-24 and Millicell®-96

24 Well and 96 Well Cell Culture Plates

Introduction

The Millicell®-24 and Millicell®-96 well cell culture plates are designed to support cell attachment, growth and differentiation for many cell applications, including transport and migration. For example, after the formation of a differentiated cell monolayer, the device can then be used to measure the rate of known and unknown drug transport across the cell barrier. All procedures are designed to be carried out in a single device and can be performed using automation for cell seeding, cell feeding, washing and experimental procedures. Both the Millicell®-24 and Millicell®-96 devices allow the cells to be visualized during the feeding and experimental stages.

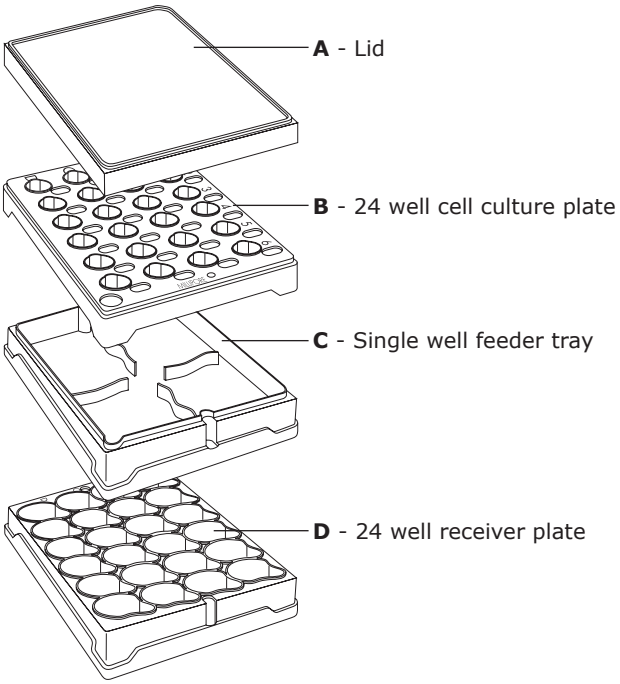
Device Storage

Store at room temperature. See expiration date on package label.

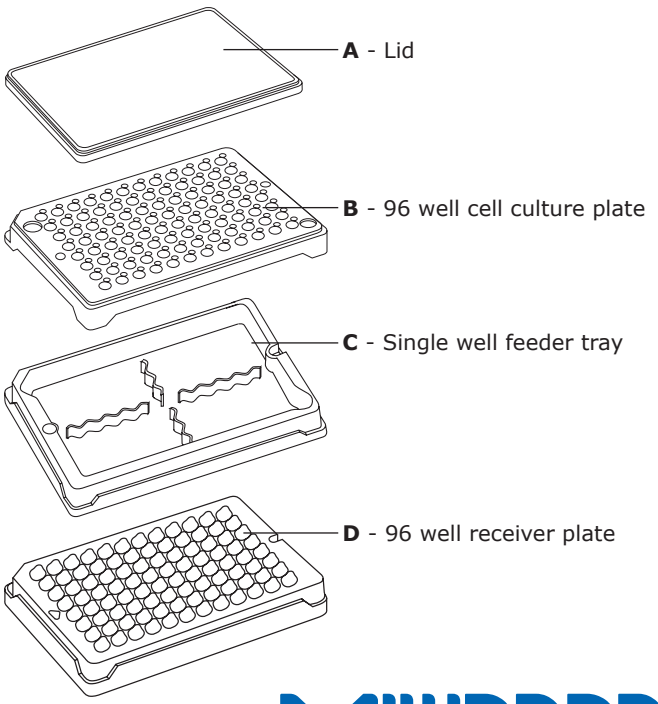
Recommended Seeding Densities

The Millicell® plates are designed to support suspension and adherent cell growth and differentiation. It is highly recommended to optimize the seeding density of the applicable cell line by exploring a range of seeding densities. Seeding densities on both devices should be similar in terms of cell count per surface area. For example, the seeding density of a 21-day culture of Caco-2 cells is approximately 80,000–86,000 cells per cm^2 of membrane. This works out to 60,000 cells/well in the Millicell®-24 device (using 400 µL/well) and 9,000 cells/well in the Millicell®-96 device (using 75 µL/well).

Millicell®-24 Plate Components



Millicell®-96 Plate Components



Seeding and Growing Cells in Millicell®-24 or Millicell®-96 Cell Culture Plates

All steps must be performed using standard cell culture aseptic technique.

1. After counting the cells, determine the appropriate seeding densities for the filter plate wells. See "Recommended Seeding Densities" on previous page.
2. Remove the lid from the assembly and add cell solution to filter plate wells while resting the tip of the aspirator above the apical assist feature inside each well.

   For Millicell®-24 plates: Add 400 µL of cell solution per well.

   For Millicell®-96 plates: Add 75 µL of cell solution per well.
3. Add cell growth medium to the single-well feeder tray or receiver plate using the basolateral access hole at the corner of the filter plate.

   For Single well feeder trays: Add 22–28 mL of cell growth medium.

   For 24 well receiver plates: Add 800 µL of cell growth medium.

   For 96 well receiver plates: Add 250 µL of cell growth medium.
4. Incubate at 37 °C, 5–6% CO_2, 95% humidity.

   NOTE: Use care to avoid tilting the plate and spilling the media when moving the plate in and out of the incubator.

Feeding Cells

When feeding cells, aspirating the volume in the single-well feeder tray using the basolateral access hole in the filter plate is recommended. Then, aspirate the medium from the filter plate wells while resting the tip of the aspirator above the apical assist feature inside each well. **Use care to avoid contacting the filter inside the wells when removing or adding medium.** Add back growth medium to the filter plate wells first (at the apical assist). Then, add cell medium to the single well feeder tray using the basolateral access hole.

It is also possible to feed the plates by separating the filter and receiver/feeder plates and utilizing the filter plate "feet." This feature allows you to place the filter plate and lid down on a solid sterile surface without any contamination. When using the Millicell®-24 plate, allow the plate assembly to rest for 15 seconds after removing it from the incubator to minimize media movement in the single-well feeder tray.

Specifications

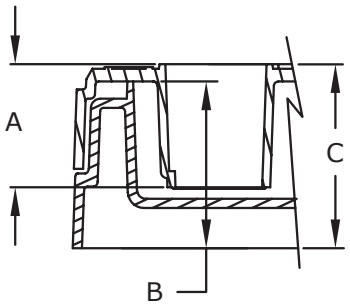
Materials of Construction	**24 Well Plate**	**96 Well Plate**
Plate, tray, and lid	Clear polystyrene	Clear polystyrene
Membrane	Polycarbonate or PET	Polycarbonate or PET
Growth/Transport Assembly		
Length × width	127.76 mm	127.76 mm
Width	85.48 mm	85.48 mm
Height of plate assembly	20.32 mm	20.32 mm
Active membrane area	0.7 cm^2	0.11 cm^2
Capacity		
Filter wells	Max. 800 µL	Max. 200 µL
24 well receiver plate	Max. 1100 µL	--
96 well receiver plate	--	Max. 400 µL
Typical Operating Volume		
Filter wells	400 µL	75 µL
24 well receiver plate	800 µL	--
96 well receiver plate	--	250 µL
Single well feeder tray	22–28 mL	24–32 mL

Properties

These sterile devices are tissue culture treated (filter plates only) and gamma irradiated (all components), ready for use as received. Extra cellular matrix (ECM) coating is not typically required for cell adhesion, but may be added for cell-specific requirements.

Automation Specifications

Plates and components are compatible with manual or robotic handling. When using a plate stacker, place an extra plate on top of the stack to ensure all plates are dispensed evenly.



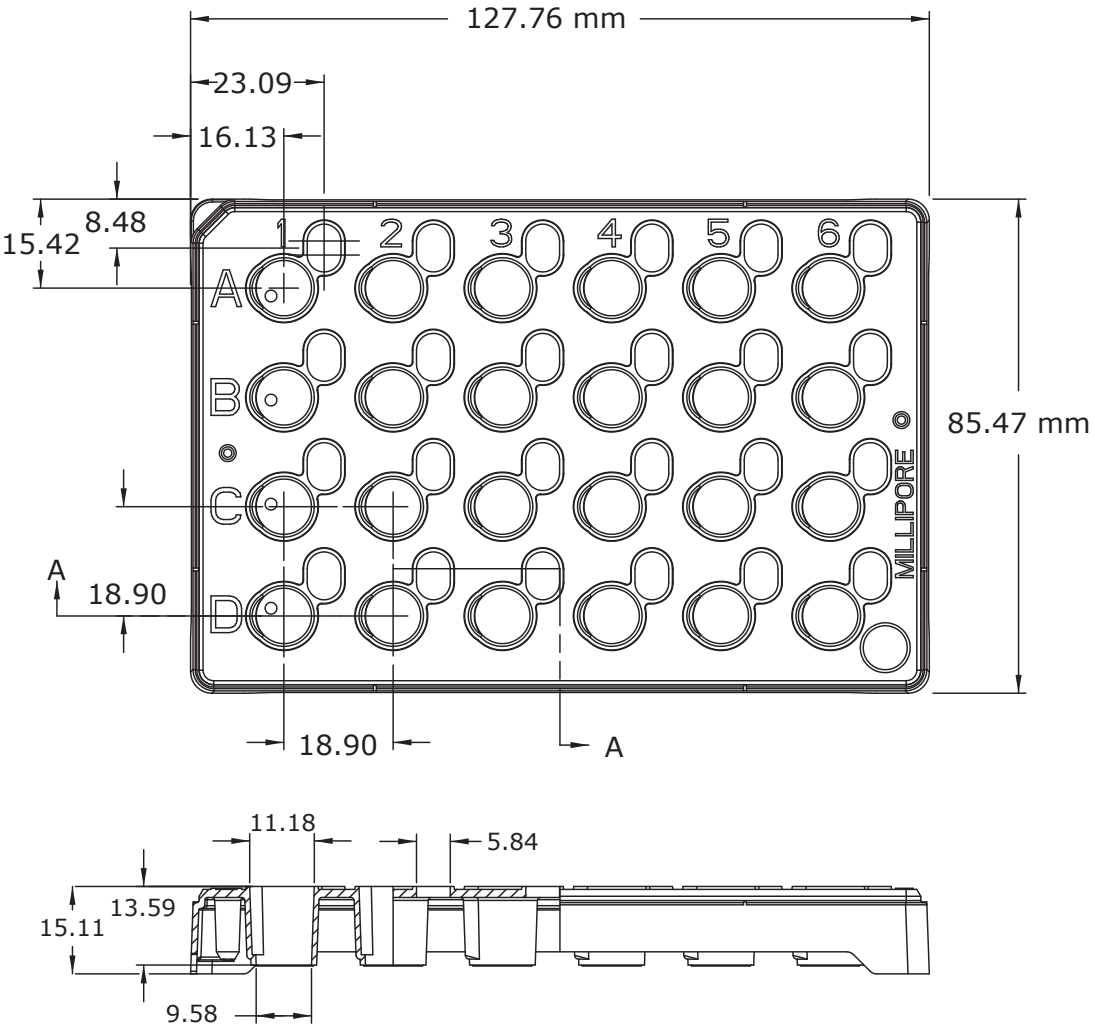
Total Assembly Dimensions

The figure below represents the dimensions in millimeters of the assembled 24 and 96 well Millicell® cell culture devices.

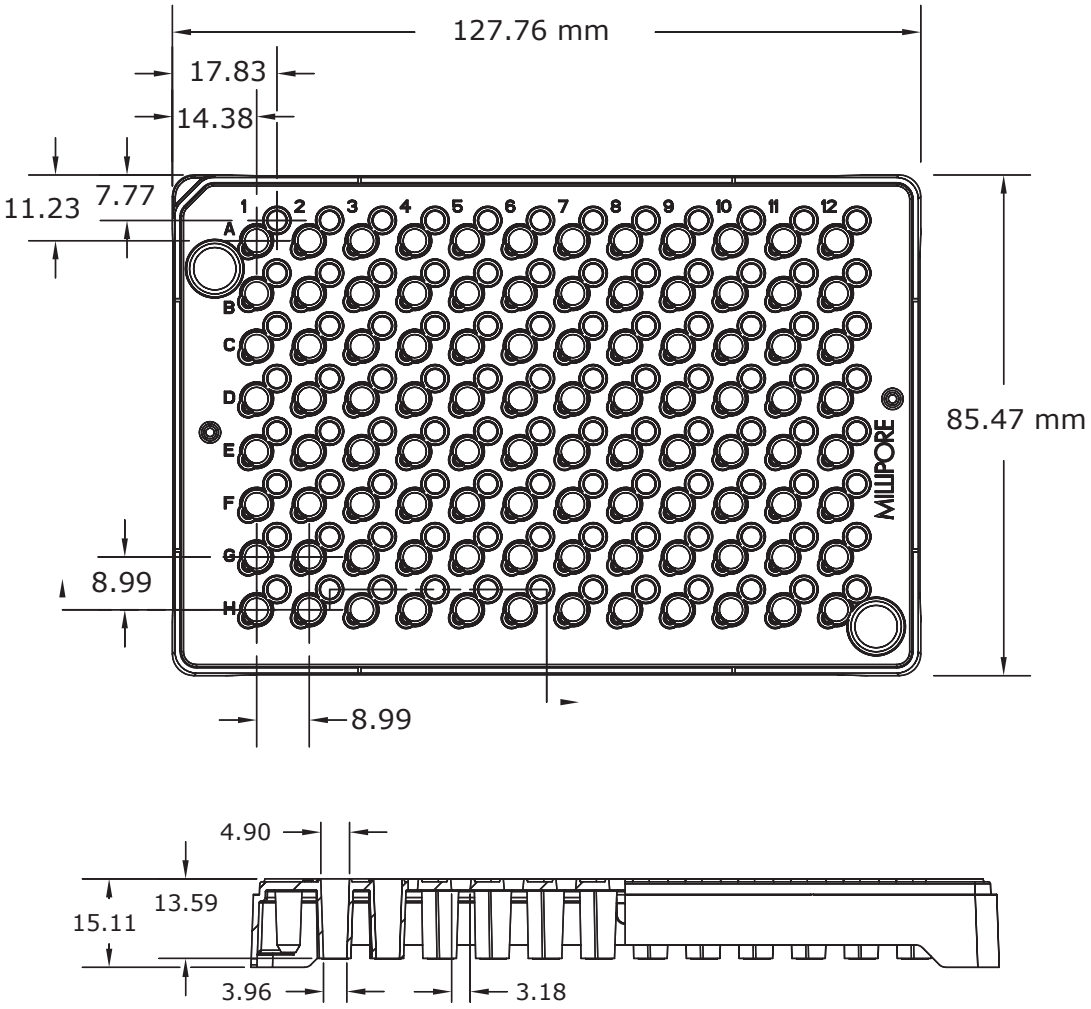
Detailed Dimensions of Millicell® Cell Plates and Lids

Key	Dimension	24 Well Plate	96 Well Plate
	Apical Well		
	A1 offset to the side	16.13 mm	14.38 mm
	A1 offset to the top	15.42 mm	11.23 mm
A	Well Depth	13.59 mm	13.59 mm
	Well Diameter — Top	11.18 mm	4.90 mm
	Well Diameter — Bottom	9.58 mm	3.96 mm
B	Receiver Plate Height (single or multi-well)	18.42 mm	18.42 mm
C	Height of Assembly (filter plate and receiver plate)	20.32 mm	20.32 mm
	Filter Plate Length	127.76 mm	127.76 mm
	Filter Plate Width	85.47 mm	85.47 mm
	Filter Plate Height	15.11 mm	15.11 mm
	Well Spacing	18.90 mm	8.99 mm
	Basolateral Well		
	A1 offset to the side	23.09 mm	17.83 mm
	A1 offset to the top	8.48 mm	7.77 mm
	Well Diameter	5.84 mm	3.18 mm
	Well depth (filter plate and single-well feeder tray)	14.86 mm	14.61 mm
	Well depth (filter plate and multi-well receiver plate)	14.99 mm	14.61 mm
	Cell Plate Lids		
	Lid Length	127.64 mm	127.64 mm
	Lid Width	85.34 mm	85.34 mm
	Lid Height	8.51 mm	8.51 mm

Detailed Dimensions of Millicell®-24 Cell Culture Plate



Detailed Dimensions of Millicell®-96 Cell Culture Plate



Product Ordering Information

Purchase products online at SigmaAldrich.com/products.

Millicell®-24 Cell Culture Plates

Components Included	Pore Size	Membrane	Qty/Pk	Cat. No.
• 24 well cell culture plate (1) • Single well feeder tray (1) • 24 well receiver plate (1) • Lid (2)	0.4 µm	PCF	1/pk	PSHT010R1
	3 µm	PCF	1/pk	PSST010R1
	5 µm	PCF	1/pk	PSMT010R1
	8 µm	PCF	1/pk	PSET010R1
	1 µm	PET	1/pk	PSRP010R1
• 24 well cell culture plate (5) • Single well feeder tray (5) • Lid (5)	0.4 µm	PCF	5/pk	PSHT010R5
	1 µm	PET	5/pk	PSRP010R5
• 24 well cell culture plate (5) • 24 well receiver plate (5) • Lid (5)	3 µm	PCF	5/pk	PSST010R5
	5 µm	PCF	5/pk	PSMT010R5
	8 µm	PCF	5/pk	PSET010R5

Accessories

24 well receiver plate (5), lid (5)	5/pk	PSMW010R5
Single well feeder tray (5), lid (5) *(for use with 24-well cell culture plate only)*	5/pk	PSSW010R5

Millicell®-96 Cell Culture Plates

Components Included	Pore Size	Membrane	Qty/Pk	Cat. No.
• 96 well cell culture plate (1) • Single well feeder tray (1) • 96 well receiver plate (1) • Lid (2)	0.4 µm	PCF	1/pk	PSHT004R1
• 96 well cell culture plate (1) • Single well feeder tray (1) • 96 well receiver plate (1) • Lid (2)	1 µm	PET	1/pk	PSRP004R1
• 96 well cell culture plate (5) • Single well feeder tray (5) • Lid (5)	0.4 µm	PCF	5/pk	PSHT004R5
• 96 well cell culture plate (5) • Single well feeder tray (5) • Lid (5)	1 µm	PET	5/pk	PSRP004R5
• 96 well cell culture plate (5) • 96 well receiver plate (10) • Lid (5)	0.4 µm	PCF	5/pk	PSHT004S5

Accessories

96 well receiver plate (5), Lid (5)	5/pk	MACAC0RS5

Additional Equipment

Millicell®-ERS-2 (Electrical Resistance System)	5/pk	MERS00002
Stericup® Quick Release GP 0.22 µm Vacuum filtration system (sterile, 500 mL)	12/pk	S2GPU05RE
Millex®-GP 0.22 µm filter unit, 33 mm	50/pk	SLGP033RS
ECL Cell Attachment Matrix (entactin-collagen IV-laminin)	5 mg	08-110

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