Microscopy Tools I: HybriWell™ Hybridization Sealing Systems, SecureSeal™ Hybridization Chambers, and HybriSlip™ Hybridization Covers

Table 1. Tools for hybridization experiments.

Product	Chamber Dimensions	Depth	Usable Volume	Quantity per Package
Fluorescent Friendly HybriWell™ hybridization sealing system				
F24724	22 mm × 22 mm	0.25 mm	30–50 µL	100
F24725	40 mm × 22 mm	0.25 mm	180–200 µL	100
F24728	60 mm × 22 mm	0.25 mm	280–330 µL	100
F24729	40 mm × 21 mm	0.15 mm	50–100 µL	100
HybriWell™ hybridization sealing system				
H24720	13 mm diameter	0.25 mm	30 µL	100
H24721	20 mm diameter	0.15 mm	30 µL	100
H24723	22 mm × 22 mm	0.15 mm	30–50 µL	100
H18210	40 mm × 21 mm	0.15 mm	50–100 µL	100
H24722	40 mm × 22 mm	0.25 mm	180–200 µL	100
SecureSeal™ hybridization chambers				
S24734	22 mm × 22 mm	0.8 mm	350 µL	50
S24730	20 mm diameter	0.8 mm	200 µL	40
S24731	20 mm diameter	1.3 mm	280 µL	40
S24732	9 mm diameter	0.8 mm	40 µL	20
S24733	9 mm diameter	1.3 mm	60 µL	20
HybriSlip™ hybridization covers				
H18200	22 mm × 22 mm	NA	NA	500
H18201	40 mm × 22 mm	NA	NA	500
H18202	60 mm × 22 mm	NA	NA	500
Seal tabs				
A18211	Adhesive seal tab	NA	NA	400

NA = Not applicable.

Introduction

Life Technologies, in association with Grace Bio-Labs, offers exciting new tools for microscopy and imaging applications. HybriWell™ hybridization sealing systems and SecureSeal™ hybridization chambers are specially designed for hybridization experiments. These ready-to-use hybridization gaskets have a special adhesive that bonds to glass slides in seconds, creating a water-tight seal that is temperature resistant, but can also be removed cleanly and easily after hybridization. The hydrophobic surfaces are nuclease-free and do not trap or bind probes like glass surfaces. Access ports in the chamber surface allow for the addition or removal of solutions and are easily sealed using seal tabs to create leak-proof chambers that eliminate evaporation. Adhesive seal tabs are provided with the HybriWell™ hybridization sealing system and the SecureSeal™ hybridization chambers, and are also available separately (Cat. no. A18211).

Table 1 provides the catalog numbers and dimensions for the various configurations of these products.

Fluorescent Friendly HybriWell™ Hybridization Sealing Systems

Fluorescent Friendly HybriWell™ hybridization sealing systems are ready-to-use hybridization gaskets that bond to glass creating a water-tight seal with access ports on the chamber surface for the addition or removal of solutions. The shallow chamber holds small volumes to minimize the amount of valuable probe used. These hydrophobic gaskets are nuclease-free, temperature resistant; and do not quench fluorescence. The fluorescent friendly versions work well with longer wavelength stains without quenching.

HybriWell™ Hybridization Sealing Systems

The HybriWell™ hybridization sealing systems (Figure 1A) are ideal for fluorescence *in situ* hybridization (FISH) to chromosomes. They create shallow chambers that hold a small reagent volume to minimize the amount of valuable probe used.

SecureSeal™ Hybridization Chambers

The SecureSeal™ hybridization chambers (Figure 1B) are larger volume chambers that provide optimum surface:volume fluid dynamics to facilitate uniform hybridization where the labeled nucleic acid is limiting, such as in microarray experiments.

HybriSlip™ Hybridization Covers

If you prefer to use coverslips for your hybridization experiments, the HybriSlip™ hybridization covers (Figure 1C) are ideally suited for *in situ* hybridization, *in situ* PCR and hybridization to genomic arrays on glass slides. Like the hybridization chambers, they are made of hydrophobic material that does not bind to nucleic acids. Because HybriSlip™ covers remain flat, even at high temperatures, they facilitate uniform reagent distribution. The HybriSlip™ covers are ready to use without pretreatment; they arrive RNase-free and protected with clean-release liners to prevent RNase contamination.

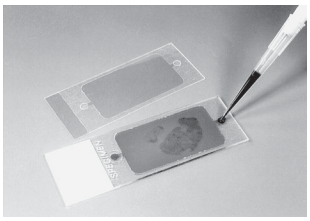

Figure 1A. HybriWell™ hybridization sealing system.

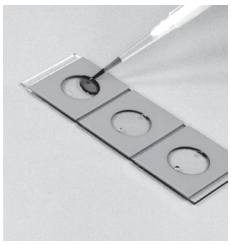

Figure 1B. SecureSeal™ hybridization chambers.

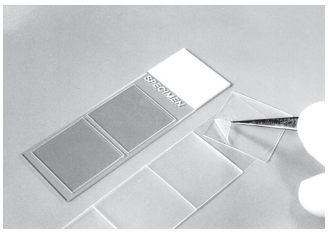

Figure 1C. HybriSlip™ hybridization covers.

How to Use

HybriWell™ Hybridization Sealing Systems

1.1 Using sterile forceps, peel off the printed liner (Figure 2A), and place the HybriWell™ seal on a flat dry surface adhesive-side up. Liner may be easily separated, starting from the tab end.

Note: HybriWell™ chambers are RNase-free and working surfaces are protected from RNase contamination by the removable liner. Take care not to contaminate exposed working surfaces.

1.2 Place a microscope slide specimen-side down over the chamber surface, aligning the edges of the slide with the edges of the HybriWell™ chamber (Figure 2B).

1.3 To ensure a secure seal, press the surface of the cover over the adhesive area with the flat edge of the smoothing tool provided.

1.4 To fill the HybriWell™ chamber, return the slide to the upright position and pipet the reagent solution through one port on the HybriWell™ seal cover while allowing air to escape through the other port (Figure 2C).

Note: For best results, we recommend that even after the chamber appears full, continue to apply the reagent until the solution is no longer drawn into the chamber. Light pressure applied to the surface of the chamber during filling dislodges air bubbles if they occur. Viscous reagents may be pipetted directly onto the surface of the HybriWell™ chamber before the microscope slide is applied.

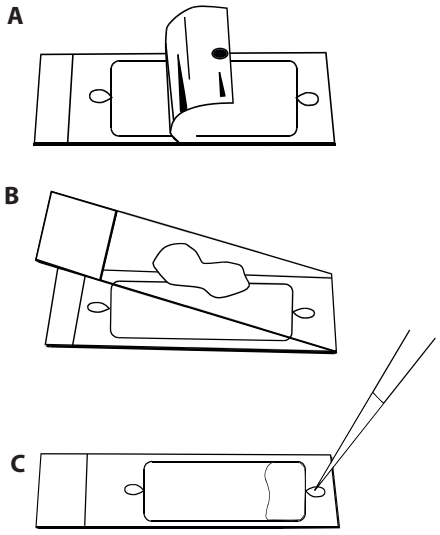

Figure 2. HybriWell™ hybridization sealing system.

1.5 To seal the ports, first wipe away excess reagent on the surface of the chamber using a tissue. Use sterile forceps to remove a seal-tab from the liner strip. Place a seal-tab over each filling port. Press on both seals simultaneously, using finger pressure, to assure a secure seal.

Note: Ports may also be sealed by applying a droplet of oil over each port.

1.6 To remove the HybriWell™ cover, grasp the tab end along the top edge of the HybriWell™ seal and peel it away from the microscope slide.

Secure-Seal™ Hybridization Chambers

2.1 Using sterile forceps, peel off the liner (Figure 3A) and place the gasket, adhesive side down, on a slide or coverslip (Figure 3B).

Note: HybriWell™ chambers are RNase-free and working surfaces are protected from RNase contamination by the removable liner. Take care not to contaminate exposed working surfaces.

2.2 To fill the SecureSeal™ chamber, pipet the reagent solution through one port on the HybriWell™ seal cover while allowing air to escape through the other port (Figure 3C).

2.3 To seal the ports with seal tabs (Cat. no. A18211, available separately), first wipe away excess reagent on the surface of the chamber using a tissue. Use sterile forceps to remove a seal-tab from the liner strip. Place a seal-tab over each filling port (Figure 3D). Press on both seals simultaneously, using finger pressure, to assure a secure seal.

Note: Ports may also be sealed by applying a droplet of oil over each port.

2.4 To remove the gasket after hybridization, simply peel it off the slide using forceps.

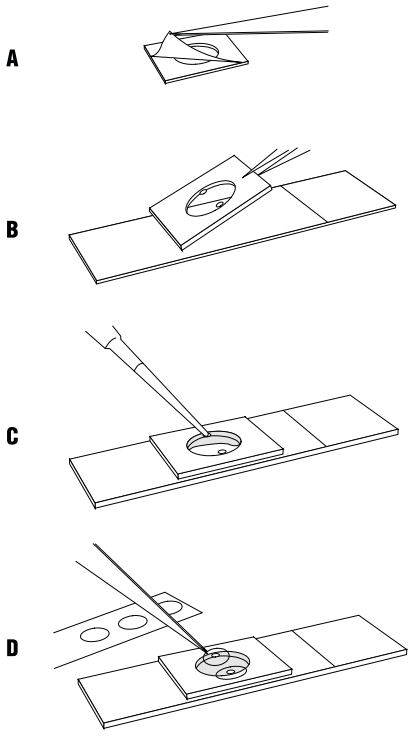

Figure 3. Secure-Seal™ hybridization chambers.

3.1 Using sterile forceps, and taking suitable precautions to guard against nuclease contamination, remove the polyethylene liner to expose the surface of the cover (Figure 4A).

3.2 Lift the HybriSlip™ cover from the support liner (Figure 4B).

3.3 Apply to specimen (Figure 4C).

3.4 Alternatively, the reagent solutions may be applied to the HybriSlip™ cover, and the specimen, inverted on a slide, applied from above. Using this approach, it is only necessary to remove the protective liner from one surface of the cover.

3.5 For high temperature applications, such as *in situ* PCR, seal the HybriSlip™ cover with nail polish before heating. Also, to avoid bubble formation, be sure to degas all reagents and warm specimens to room temperature before use.

3.6 To remove the HybriSlip™ cover, first cut through the seal with a razor blade, if sealed, and then apply buffer to float off the cover.

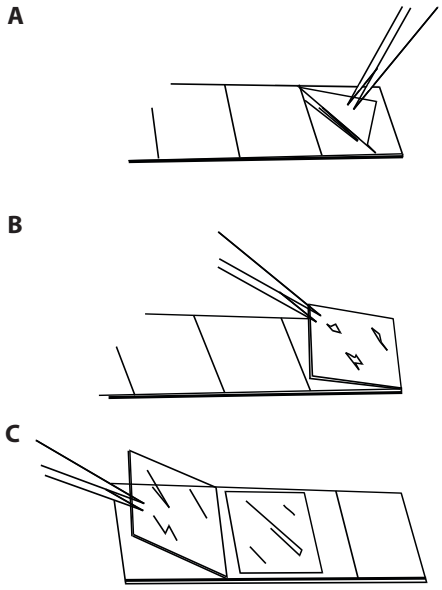

Figure 4. HybriSlip™ hybridization covers.

Product List Current prices may be obtained from our website or from our Customer Service Department.

Cat. no.	Product Name	Unit Size
A18211	Adhesive seal-tab, for HybriWell™ hybridization sealing system *set of 400*	1 set
F24724	Fluorescent Friendly HybriWell™ hybridization sealing system, 22 mm x 22 mm chamber, 0.25 mm deep HBW2222-FL *set of 100*	1 set
F24725	Fluorescent Friendly HybriWell™ hybridization sealing system, 40 mm x 22 mm chamber, 0.25 mm deep HBW2240-FL *set of 100*	1 set
F24728	Fluorescent Friendly HybriWell™ hybridization sealing system, 60 mm x 22 mm chamber, 0.25 mm deep HBW2260-FL *set of 100*	1 set
F24729	Fluorescent Friendly HybriWell™ hybridization sealing system, 40 mm x 22 mm chamber, 0.15 mm deep HBW75-FL *set of 100*	1 set
H18200	HybriSlip™ hybridization cover, 22 mm x 22 mm *RNase free* *set of 500*	1 set
H18201	HybriSlip™ hybridization cover, 40 mm x 22 mm *RNase free* *set of 500*	1 set
H18202	HybriSlip™ hybridization cover, 60 mm x 22 mm *RNase free* *set of 500*	1 set
H24720	HybriWell™ hybridization sealing system, 13 mm diameter chamber, 0.25 mm deep *set of 100*	1 set
H24721	HybriWell™ hybridization sealing system, 20 mm diameter chamber, 0.15 mm deep *set of 100*	1 set
H24723	HybriWell™ hybridization sealing system, 22 mm x 22 mm chamber, 0.15 mm deep *set of 100*	1 set
H18210	HybriWell™ hybridization sealing system, 40 mm x 21 mm chamber, 0.15 mm deep *set of 100*	1 set
H24722	HybriWell™ hybridization sealing system, 40 mm x 22 mm chamber, 0.25 mm deep *set of 100*	1 set
S24734	SecureSeal™ hybridization chamber gasket, one chamber, 22 mm x 22 mm, 0.8 mm deep *set of 50*	1 set
S24730	SecureSeal™ hybridization chamber gasket, one chamber, 20 mm diameter, 0.8 mm deep *set of 40*	1 set
S24731	SecureSeal™ hybridization chamber gasket, one chamber, 20 mm diameter, 1.3 mm deep *set of 40*	1 set
S24732	SecureSeal™ hybridization chamber gasket, eight chambers, 9 mm diameter, 0.8 mm deep *set of 20*	1 set
S24733	SecureSeal™ hybridization chamber gasket, eight chambers, 9 mm diameter, 1.3 mm deep *set of 20*	1 set

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