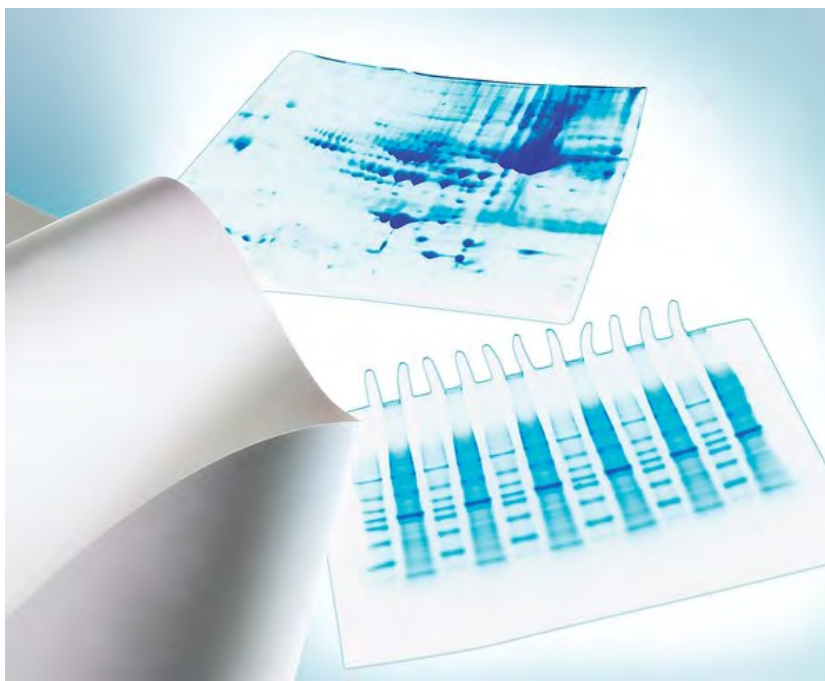


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**Россия** +7(495)268-04-70

**Казахстан** +7(7172)727-132

**Киргизия** +996(312)96-26-47

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# Immobilon™ Transfer Membranes

*For superior protein and nucleic acid blots*

Membranes for:

- ▶ Westerns
- ▶ Dots/slots
- ▶ Southern
- ▶ Northern
- ▶ Colony/plaque lifts

## Protein Applications

### Immobilon-P Membrane

The original, and most commonly used, PVDF transfer membrane for western blotting. It has a 0.45 µm pore size and is recommended for blotting proteins >20 kDa.

- Compatible with a variety of detection chemistries, including radioactive, chromogenic, and chemiluminescent techniques
- High protein adsorption and retention ensures greater sensitivity
- Won't crack or curl like nitrocellulose. Can be cut without fracturing and reprobed multiple times
- Our Rapid Immunodetection Protocol eliminates the need for blocking and reduces detection times by up to 2 hours

### Immobilon-PSQ Membrane

This PVDF membrane has a 0.2 µm pore size. The large internal structure results in higher protein adsorption and sequencing yields than other membranes. Recommended for blotting proteins <20 kDa and sequencing.

- Higher capacity and retention than 0.45 µm membranes
- Prevents blow-through of low molecular weight proteins
- Compatible with a variety of detection chemistries, including radioactive, chromogenic, and chemiluminescent techniques

### Immobilon-FL Membrane

The first transfer membrane optimized for fluorescence applications. This PVDF membrane exhibits extremely low background fluorescence.

- Compatible with all commonly used fluorescent dyes
- Can be used at all excitation and emission wavelengths. Ideal for multiplexing
- Quality tested to ensure low background fluorescence in western blotting applications
- Also compatible with non-fluorescent detection chemistries

## Nucleic Acid Applications

### Immobilon-Ny+ Membrane

A positively charged nylon membrane optimized for reliable and reproducible transfer, immobilization, hybridization, and subsequent reprobing.

- Provides maximum sensitivity with minimal background due to the density and uniformity of the positively charged surface
- Has exceptional retention and reprobing characteristics. Studies show 50% greater signal than other positively charged nylon membranes—5x greater after 12 reprobes
- Performs well in both chemiluminescent and radioactive detection systems
- Can be used to detect sub-picrogram amounts of DNA and RNA

### Immobilon-NC Membranes

An economical alternative for nucleic acid and protein blotting protocols.

- Immobilon-NC (HAHY) membrane is a mixed cellulose esters matrix with surfactants that improve wettability and handling during the transfer process.
- Immobilon-NC (HATF) membrane is a mixed cellulose esters matrix with no surfactants that can interfere with cell wall integrity during cell growth. Recommended for colony lifts.

## Blotting Sandwiches with Immobilon-P Membrane

- Immobilon-P membrane interleaved with two sheets of pre-cut blotting paper offers convenience and time savings for high throughput labs. Available in sizes to match most pre-cut gels. See Ordering Information for cut sizes.

## Selecting an Immobilon Membrane for Your Application

|                                  | Immobilon-P                                                                                                                                                                                                                                                                                                   | Immobilon-PSQ                                                                                                                                                                                  | Immobilon-FL                                                                                                                              | Immobilon-NY+                                                                                                                                                                                                                                                            | Immobilon-NC (HAHY)                                                                                                            |
|----------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------|
| Description                      | The original PVDF membrane for western blotting applications                                                                                                                                                                                                                                                  | Small pore structure results in superior binding of proteins with MW <20 kDa                                                                                                                   | Optimized for fluorescent-based immunodetection applications                                                                              | Optimized for general nucleic acid applications                                                                                                                                                                                                                          | Optimized for RNA and DNA blotting applications                                                                                |
| Composition                      | PVDF<br>Blotting sandwiches include pre-cut membrane and two sheets of blotting paper                                                                                                                                                                                                                         | PVDF                                                                                                                                                                                           | PVDF                                                                                                                                      | Positively charged nylon with polyester reinforcement                                                                                                                                                                                                                    | Mixed cellulose esters with surfactants to improve wettability and handling                                                    |
| Pore size                        | 0.45 µm                                                                                                                                                                                                                                                                                                       | 0.2 µm                                                                                                                                                                                         | 0.45 µm                                                                                                                                   | 0.45 µm                                                                                                                                                                                                                                                                  | 0.45 µm                                                                                                                        |
| Phobicity                        | Hydrophobic                                                                                                                                                                                                                                                                                                   | Hydrophobic                                                                                                                                                                                    | Hydrophobic                                                                                                                               | Hydrophilic                                                                                                                                                                                                                                                              | Hydrophilic                                                                                                                    |
| Applications                     | <ul style="list-style-type: none"> <li>• Western blotting</li> <li>• Binding assays</li> <li>• Amino acid analysis</li> <li>• N-terminal protein sequencing</li> <li>• Dot/slot blotting</li> <li>• Glycoprotein visualization</li> <li>• Lipopolysaccharide analysis</li> <li>• Mass spectrometry</li> </ul> | <ul style="list-style-type: none"> <li>• Low molecular weight western blotting</li> <li>• Amino acid analysis</li> <li>• Mass spectrometry</li> <li>• N-terminal protein sequencing</li> </ul> | <ul style="list-style-type: none"> <li>• Western blotting</li> <li>• Dot/slot blotting</li> <li>• Fluorescence-based detection</li> </ul> | <ul style="list-style-type: none"> <li>• Southern blotting</li> <li>• Northern blotting</li> <li>• Nucleic acid reprobing</li> <li>• Gene arrays</li> <li>• DNA finger-printing</li> <li>• Colony lifts</li> <li>• Plaques lifts</li> <li>• NA dot/slot blots</li> </ul> | <ul style="list-style-type: none"> <li>• Southern blotting</li> <li>• Northern blotting</li> <li>• Western blotting</li> </ul> |
| Detection methods                | Chemiluminescent<br>Chromogenic<br>Radioactive                                                                                                                                                                                                                                                                | Chemiluminescent<br>Chromogenic<br>Radioactive<br>Fluorescent                                                                                                                                  | Fluorescent<br>Chemifluorescent<br>Chromogenic<br>Chemiluminescent                                                                        | Chemiluminescent<br>Radioactive                                                                                                                                                                                                                                          | Chemiluminescent<br>Radioactive                                                                                                |
| Typical protein binding capacity | Insulin: 85 µg/cm <sup>2</sup><br>BSA: 131 µg/cm <sup>2</sup><br>Goat IgG: 294 µg/cm <sup>2</sup>                                                                                                                                                                                                             | Insulin: 262 µg/cm <sup>2</sup><br>BSA: 340 µg/cm <sup>2</sup><br>Goat IgG: 448 µg/cm <sup>2</sup>                                                                                             | Insulin: 85 µg/cm <sup>2</sup><br>BSA: 131 µg/cm <sup>2</sup><br>Goat IgG: 294 µg/cm <sup>2</sup>                                         | Not applicable                                                                                                                                                                                                                                                           | Not applicable                                                                                                                 |
| Compatible stains                | Coomassie™ Brilliant Blue<br>Amido black<br>India ink<br>Ponceau-S red<br>Colloidal gold<br>CPTS<br>Toluidine blue<br>Transillumination<br>Sypro® Ruby blot stain                                                                                                                                             | Coomassie Brilliant Blue<br>Amido black<br>India ink<br>Ponceau-S red<br>Colloidal gold<br>CPTS<br>Toluidine blue<br>Transillumination<br>Sypro Ruby blot stain                                | Sypro Ruby blot stain<br>Coomassie Brilliant Blue<br>Amido black<br>Ponceau-S red<br>CPTS<br>Transillumination                            | Not applicable                                                                                                                                                                                                                                                           | Not applicable                                                                                                                 |

## Performance

### Immobilon-P Membrane (0.45 µm)

Optimized for Superior Western Blots (chemiluminescent or chromogenic)

#### Immobilon-NC (HATF)

Detergent-free  
membrane for colony  
and plaque lifts

Mixed cellulose  
esters without  
surfactants

0.45 µm

Hydrophilic

- Colony lifts
- Plaque lifts

Radioactive

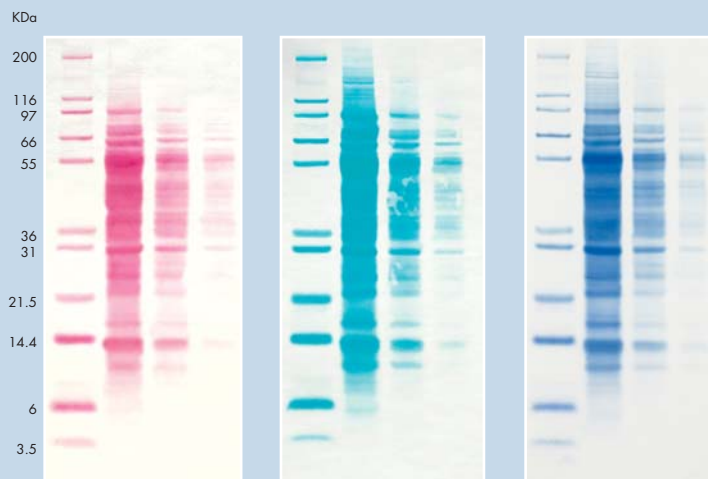
Not applicable

Not applicable

#### Rapid Immunodetection Method Reduces Blotting Time

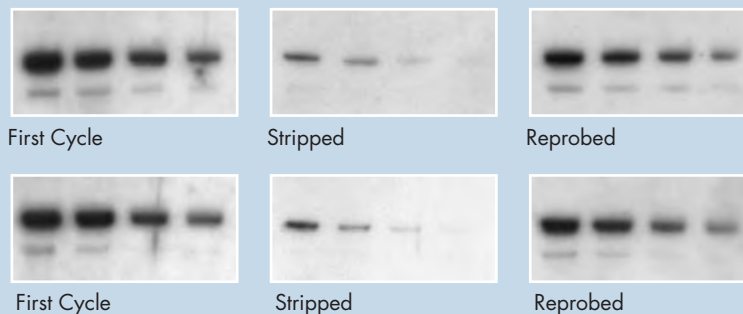
| Step                                   | Standard<br>Immunodetection | Rapid<br>Immunodetection |
|----------------------------------------|-----------------------------|--------------------------|
| 1. Block the membrane                  | 1 hr                        | None                     |
| 2. Incubate with primary<br>antibody   | 1 hr                        | 1 hr                     |
| 3. Wash the membrane                   | 3 x 10 min                  | 3 x 5 min                |
| 4. Incubate with secondary<br>antibody | 1 hr                        | 30 min                   |
| 5. Wash the membrane                   | 3 x 10 min                  | 3 x 5 min                |
| 6. Add substrate                       | 10 min                      | 10 min                   |
| Total time                             | 4 hr 10 min                 | 2 hr 10 min              |

#### Compatible with All Commonly Used Stains



Western blots of calf liver  
proteins detected with (left to  
right): Ponceau-S red, CPTS,  
and Coomassie Brilliant Blue  
total protein blot stains.  
Lanes (left to right): molecular  
weight standards, 20, 5,  
and 1.25 µg of liver  
proteins.

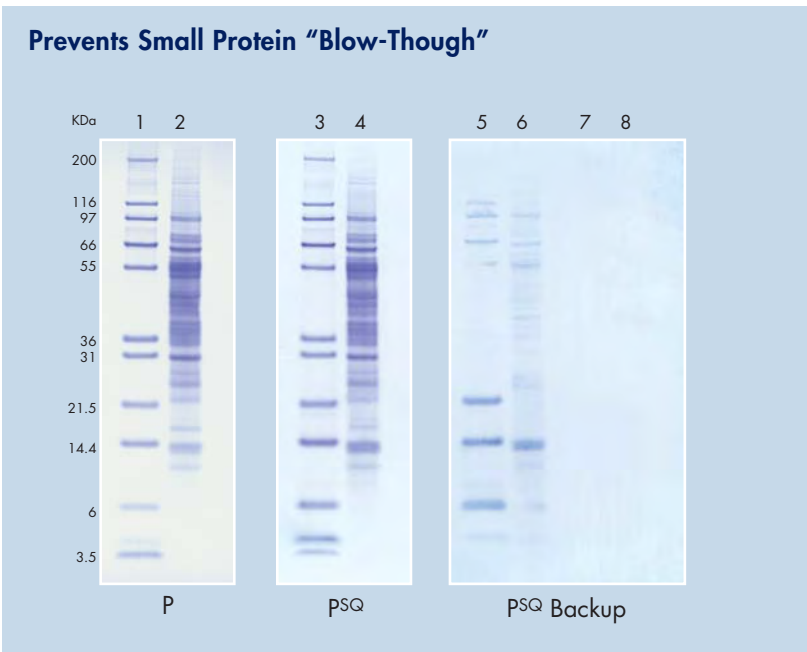
#### Superior Reprobing with Detergent and Low pH Methods



Reprobing Immobilon-P mem-  
brane with anti-human transferrin  
(1:10,000 dilution) and HRP-  
conjugated rabbit anti-goat IgG  
(1:20,000) by detergent (top  
row) and low pH methods  
(bottom row) using ECL  
(Amersham) detection reagents.  
(1) First cycle of detection;  
(2) stripped membrane detected  
with the secondary antibody  
only; (3) second cycle—  
stripped membrane detected  
with primary and secondary  
antibody. Left to right, 5 µL of  
human serum dilutions  
1:12,500, 1:25,000,  
1:50,000, and 1:100,000.

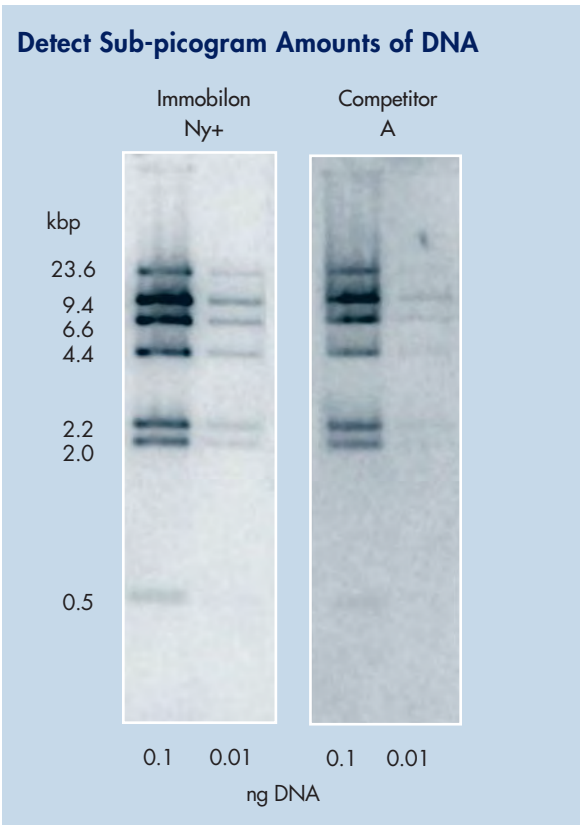
## Immobilon-PSQ Membrane (0.2 µm)

Large Internal Surface for Blotting Proteins <20 kDa and for Sequencing



## Immobilon-Ny+ Membrane (0.45 µm)

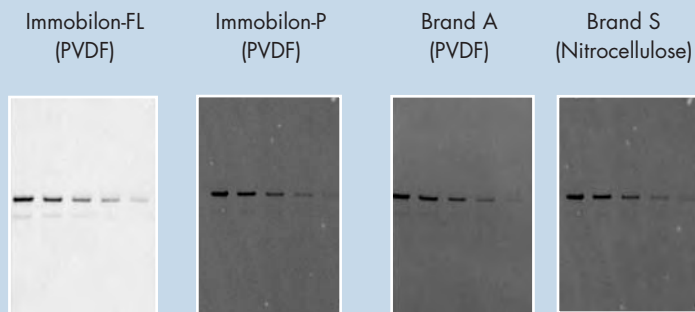
Optimized for Reliable and Reproducible Transfer, Immobilization, Hybridization, and Reprobing



## Immobilon-FL Membrane (0.45 $\mu\text{m}$ )

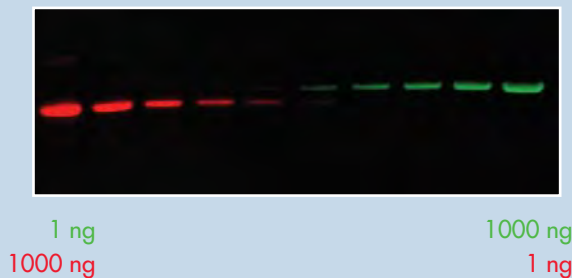
Optimized for Fluorescence-based Immunodetection

### Lower Background than Nitrocellulose or Other PVDF Membranes



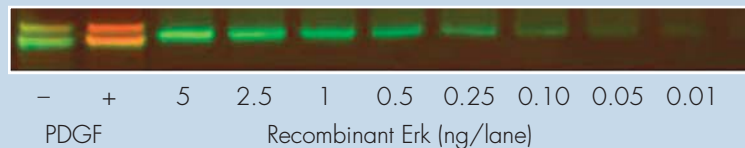
Reverse image showing fluorescent detection of transferrin in human serum using FITC-conjugated second antibody. Serum dilutions: 1:4,000, 1:8,000, 1:16,000, 1:32,000, 1:64,000. All membranes were scanned using a Storm<sup>TM</sup> 840 imaging system in blue fluorescence mode. Background fluorescence levels of Immobilon-FL membrane are up to 10X less than other blotting membranes.

### Ideal for Multiplexing



Actin-tubulin assay on Immobilon-FL membrane. Rabbit muscle actin (red) was detected using rabbit anti-actin 1<sup>o</sup>AB and QDot<sup>®</sup> 655 goat anti-rabbit 2<sup>o</sup>AB. Porcine brain tubulin (green) was detected using mouse anti-tubulin 1<sup>o</sup>AB and QDot 565 goat anti-mouse 2<sup>o</sup>AB. Sensitivities down to 1 ng were observed on a Kodak imager. Data provided by Quantum Dot Corporation.

### Quantify Both Total and Phosphorylated Protein Expression



MAP kinase assay in PDGF-treated 3T3 cells shows excellent sensitivity and multiplex labeling using Qdot 605 anti-rabbit conjugate (pan Erk) and Qdot 705 anti-mouse conjugate (phospho Erk). Bands are pseudocolored for better visualization. Data provided by Quantum Dot Corporation.

## Ordering Information

### Immobilon-P Membrane, PVDF, 0.45 µm pore size

| Type  | Size             | Qty/Pk | Catalogue No. |
|-------|------------------|--------|---------------|
| Sheet | 7 x 8.4 cm       | 50     | IPVH 078 50   |
|       | 8 x 10 cm        | 10     | IPVH 081 00   |
|       | 8.5 x 13.5       | 10     | IPVH 081 30   |
|       | 9 x 12 cm        | 10     | IPVH 091 20   |
|       | 10 x 10 cm       | 10     | IPVH 101 00   |
|       | 15 x 15 cm       | 10     | IPVH 151 50   |
|       | 20 x 20 cm       | 10     | IPVH 202 00   |
|       | 26 x 26 cm       | 10     | IPVH 304 FO   |
| Roll  | 26.5 cm x 3.75 m | 1      | IPVH 000 10   |

### Blotting Sandwiches

| Type                                                 | Size          | Qty/Pk | Catalogue No. |
|------------------------------------------------------|---------------|--------|---------------|
| Immobilon-P membrane interleaved with blotting paper | 7 x 8.4 cm    | 20     | IPSN 078 52   |
|                                                      | 8.5 x 13.5 cm | 20     | IPSN 081 32   |

### Immobilon-PSQ Membrane, PVDF, 0.2 µm pore size

| Type  | Size             | Qty/Pk | Catalogue No. |
|-------|------------------|--------|---------------|
| Sheet | 7 x 8.4 cm       | 10     | ISEQ 078 50   |
|       | 8 x 10 cm        | 10     | ISEQ 081 00   |
|       | 8.5 x 13.5       | 10     | ISEQ 081 30   |
|       | 9 x 12 cm        | 10     | ISEQ 091 20   |
|       | 10 x 10 cm       | 10     | ISEQ 101 00   |
|       | 15 x 15 cm       | 10     | ISEQ 151 50   |
|       | 20 x 20 cm       | 10     | ISEQ 202 00   |
|       | 26 x 26 cm       | 10     | ISEQ 262 60   |
| Roll  | 26.5 cm x 3.75 m | 1      | ISEQ 000 10   |

### Immobilon-FL Membrane, PVDF, 0.45 µm pore size

| Type  | Size             | Qty/Pk | Catalogue No. |
|-------|------------------|--------|---------------|
| Sheet | 10 x 10 cm       | 10     | IPFL 101 00   |
|       | 20 x 20 cm       | 10     | IPFL 202 00   |
| Roll  | 26.5 cm x 3.75 m | 1      | IPFL 000 10   |

### Immobilon-Ny+ Membrane, web-supported positively charged nylon, 0.45 µm pore size

| Type  | Size           | Qty/Pk | Catalogue No. |
|-------|----------------|--------|---------------|
| Disc  | 82 mm          | 50     | INYC 082 50   |
|       | 85 mm          | 50     | INYC 085 50   |
|       | 132 mm         | 50     | INYC 132 50   |
|       | 137 mm         | 50     | INYC 137 50   |
| Sheet | 6 x 11 cm      | 50     | INYC 611 50   |
|       | 9 x 12 cm      | 10     | INYC 091 20   |
|       | 10 x 10 cm     | 10     | INYC 101 00   |
|       | 15 x 15 cm     | 10     | INYC 151 50   |
|       | 20 x 20 cm     | 10     | INYC 202 00   |
|       | 22.2 x 22.2 cm | 10     | INYC 222 20   |
|       | 22.2 x 22.2 cm | 100    | INYC 222 2C   |
|       | 26 x 26 cm     | 10     | INYC 262 60   |
| Roll  | 30 cm x 3.3 m  | 1      | INYC 000 10   |

### Immobilon-NC (HATF), surfactant-free MCE, 0.45 µm pore size

| Type | Size   | Qty/Pk | Catalogue No. |
|------|--------|--------|---------------|
| Disc | 82 mm  | 50     | HATF 082 50   |
|      | 85 mm  | 50     | HATF 085 50   |
|      | 132 mm | 50     | HATF 132 50   |
|      | 137 mm | 50     | HATF 137 50   |

### Immobilon-NC (HAHY), MCE, 0.45 µm pore size

| Type  | Size        | Qty/Pk | Catalogue No. |
|-------|-------------|--------|---------------|
| Disc  | 82 mm       | 50     | HAHY 082 50   |
|       | 85 mm       | 50     | HAHY 085 50   |
|       | 137 mm      | 50     | HAHY 137 50   |
| Sheet | 30 x 30 cm  | 50     | HAHY 304 FO   |
| Roll  | 33 cm x 3 m | 1      | HAHY 000 10   |



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Россия +7(495)268-04-70

Казахстан +7(7172)727-132

Киргизия +996(312)96-26-47

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