The Blocking reagent is used to decrease the background in non-radioactive hybridization and detection of nucleic acid hybrids.

**Product overview**

**Appearance** Powder.

**Application**
The Blocking reagent is used to decrease the background in non-radioactive hybridization and detection of nucleic acid hybrids.

**Storage/ Stability**
The unopened reagent is stable at +15 to +25°C until the expiration date printed on the label. **Note:** We recommend to store the concentrated 10x Blocking solution in aliquots at -15 to -25°C.

**Preparation of 10x Blocking stock solution for filter hybridization**

**Additional required reagents** Maleic acid buffer: 100 mM Maleic acid, 150 mM NaCl, pH 7.5 (20°C), adjusted with conc. or solid NaOH, sterile.

**Preparation of Blocking stock solution 10x**

Blocking reagent is dissolved in maleic acid buffer to a final concentration of 10% (w/v) with shaking and heating either on a heating block or in a microwave oven.

This stock solution is autoclaved and stored at +2 to +8°C or -15 to -25°C subsequently.

**Preparation of 1x Blocking solution**

Dilute the 10x Blocking stock solution with 1x Maleic acid buffer to a 1x concentrated solution.

**Always prepare fresh!**

**Preparation of hybridization buffers**

For the hybridization (and pre-hybridization) of filters with DIG-labeled probes, the addition of Blocking reagent to the hybridization buffer is recommended.

**Use of Blocking reagent in buffers**

For the hybridization (and pre-hybridization) of filters with DIG-labeled probes, the addition of Blocking reagent to the hybridization buffer is recommended.

<table>
<thead>
<tr>
<th>Hybridization buffer</th>
<th>Composition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Standard hybridization buffer</td>
<td>5x SSC, 0.1% N-lauroylsarcosine (w/v), 0.02% SDS (w/v), 1% Blocking solution (v/v) (1/10 volume of Blocking solution, 10x conc.)</td>
</tr>
<tr>
<td>Standard hybridization buffer with formamide</td>
<td>50% formamide (v/v) deionized, 5x SSC, 0.1% N-lauroylsarcosine (w/v), 0.02% SDS (w/v), 2% Blocking solution (v/v) (1/5 volume of Blocking solution, 10x conc.)</td>
</tr>
<tr>
<td>High SDS hybridization buffer</td>
<td>7% SDS, 50% formamide (w/v) deionized, 5x SSC, 50 mM sodium phosphate, pH 7.0, 0.1% N-lauroylsarcosine (v/v), 2% Blocking solution (w/v) (1/5 volume of Blocking solution, 10x conc.)</td>
</tr>
</tbody>
</table>

**Hybridization conditions**

The hybridization conditions depend largely on the type of probe (DNA, RNA or oligonucleotide) and are described in detail in the working procedures of the corresponding DIG kits (see below).

Detailed working instructions and practical hints, concerning probe labeling with DIG, hybridization and chemiluminescent color detection, are described in the DIG Kits (listed below) and in the DIG Applications Guide for Filter Hybridization, available via internet or local Roche Applied Science representative.

**Ordering Information**

**Kits**

<table>
<thead>
<tr>
<th>Product</th>
<th>Pack Size</th>
<th>Cat. No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>DIG High Prime Labeling and Detection Starter Kit I</td>
<td>12 labeling reactions and 24 blots</td>
<td>11 745 832 910</td>
</tr>
<tr>
<td>DIG High Prime Labeling and Detection Starter Kit II</td>
<td>12 labeling reactions and 24 blots</td>
<td>11 685 614 910</td>
</tr>
<tr>
<td>DIG Northern Starter Kit</td>
<td>10 labeling reactions and detection</td>
<td>12 039 672 910</td>
</tr>
<tr>
<td>DIG DNA Labeling and Detection Kit</td>
<td>25 labeling reactions and 50 blots</td>
<td>11 039 672 910</td>
</tr>
<tr>
<td>DIG Nucleic Acid Detection Kit</td>
<td>40 (10x10 cm)</td>
<td>11 175 041 910</td>
</tr>
<tr>
<td>DIG Luminescent Detection Kit</td>
<td>50 (10x10 cm)</td>
<td>11 363 514 910</td>
</tr>
</tbody>
</table>

**Single reagents**

<table>
<thead>
<tr>
<th>Product</th>
<th>Pack Size</th>
<th>Cat. No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>DIG Easy Hyb</td>
<td>500 ml</td>
<td>11 603 588 001</td>
</tr>
<tr>
<td>DIG Easy Hyb Granules</td>
<td>50 ml</td>
<td>11 796 705 001</td>
</tr>
<tr>
<td>Nylon membranes, positively charged</td>
<td>10 sheets (20 x 30 cm)</td>
<td>11 209 272 910</td>
</tr>
<tr>
<td></td>
<td>20 sheets (10 x 15 cm)</td>
<td>11 209 299 910</td>
</tr>
<tr>
<td></td>
<td>1 roll (0.3 x 3 m)</td>
<td>11 417 240 910</td>
</tr>
<tr>
<td>Nylon membranes for Colony and Plaque Hybridization</td>
<td>50 discs (each 82 mm diameter)</td>
<td>11 699 075 001</td>
</tr>
<tr>
<td></td>
<td>50 discs (each 132 mm diameter)</td>
<td>11 699 083 001</td>
</tr>
<tr>
<td>Blocking reagent</td>
<td>50 g</td>
<td>11 096 176 001</td>
</tr>
<tr>
<td>Hybridization bags</td>
<td>50 bags</td>
<td>11 686 649 001</td>
</tr>
<tr>
<td>Anti digoxigenin-AP conjugate, Fab fragments</td>
<td>150 μl (200 μl)</td>
<td>11 093 274 910</td>
</tr>
<tr>
<td>NBT/BCIP stock solution tablets</td>
<td>6 ml</td>
<td>11 681 451 001</td>
</tr>
<tr>
<td>CSPD</td>
<td>1 ml</td>
<td>11 655 884 001</td>
</tr>
<tr>
<td>CSPD, ready-to-use</td>
<td>2 x 50 ml</td>
<td>11 755 633 001</td>
</tr>
<tr>
<td>CDP-Star</td>
<td>1 ml</td>
<td>11 685 672 001</td>
</tr>
<tr>
<td>CDP-Star, ready-to-use</td>
<td>2 x 50 ml</td>
<td>12 041 677 001</td>
</tr>
<tr>
<td>Tween 20</td>
<td>5 x 10 ml</td>
<td>11 332 465 001</td>
</tr>
</tbody>
</table>

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* available from Roche Applied Sciences

**Contact and Support**

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