E-Gel® EX agarose gels are pre-cast 1%, 2%, and 4% agarose gels, for use with the E-Gel® iBase™ Power System. E-Gel® EX gels have 11 wells, and a novel openable format. A proprietary fluorescent nucleic acid stain in the gel allows detection down to 1 ng/band of DNA when visualized by blue light transilluminator (excitation at 490 nm/emission at 522 nm). For more information and detailed instructions, refer to the E-Gel® Technical Guide available at www.invitrogen.com or contact Technical Support.

**E-Gel® EX Agarose Gels**

Catalog nos. G6511ST, G6512ST, G4020-01, G4020-02, G4010-01, G4010-02, G4010-04

Part no. 25-1038 Rev. Date: 6 August 2009

**General Guidelines**

- Store gels at room temperature
- For samples or DNA ladders in high salt buffer, dilute 2- to 20-fold before loading
- Load 100–250 ng of DNA ladder diluted in an appropriate volume
- Prepare DNA samples and markers in deionized water, or the E-Gel® Sample Loading Buffer (Invitrogen Catalog no. 10482-055)
- Keep sample volumes uniform and load deionized water into empty wells
- Use a blue-light transilluminator (e.g., E-Gel® Safe Imager™) to visualize DNA (observe safety instructions described in manual)
- To prepare and run RNA samples, refer to the E-Gel® Technical Guide

**Sample Preparation**

- Use a total sample volume of 20 μl for each well
- Adjust the amount of DNA sample according to the number of bands being separated

<table>
<thead>
<tr>
<th>Agarose Gel %</th>
<th>Single DNA Band</th>
<th>Multiple DNA Bands</th>
<th>Optimal Sample Amount</th>
<th>Maximum Sample Amount</th>
</tr>
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<tr>
<td>1%</td>
<td>1–100 ng</td>
<td>1–50 ng/band</td>
<td>3–25 ng</td>
<td>250 ng</td>
</tr>
<tr>
<td>2%</td>
<td>1–300 ng</td>
<td>1–100 ng/band</td>
<td>5–150 ng</td>
<td>500 ng</td>
</tr>
<tr>
<td>4%</td>
<td>1–300 ng</td>
<td>1–100 ng/band</td>
<td>5–200 ng</td>
<td>500 ng</td>
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**Troubleshooting**

<table>
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<tr>
<td>Leaking samples</td>
<td>Wells damaged during comb removal</td>
<td>Remove comb gently without damaging the wells.</td>
</tr>
<tr>
<td>High background, sub-optimal or no image</td>
<td>No filter or wrong filter set</td>
<td>Refer to E-Gel® Technical Guide or instrument manufacturer for optimal filter set.</td>
</tr>
<tr>
<td></td>
<td>Photographic settings not optimal</td>
<td>Determine optimal settings empirically by adjusting exposure time, gain, etc.</td>
</tr>
</tbody>
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**One-Step Loading of E-Gel® EX Agarose Gel**

1. If using the iBase™ without an E-Gel® Safe Imager™, connect the cord with the transformer (a) to the power inlet of the iBase™, and plug the other end into an electrical outlet. Verify that your iBase™ firmware has the following programs to run your gels:
   - “E-Gel® EX 1−2%” program for E-Gel® EX 1% and 2% gels
   - “E-Gel® EX 4%” program for 4% E-Gel® EX gels

   See [Downloading Upgrades](#) if programs are not present.

2. If using the iBase™ with an E-Gel® Safe Imager™, place the iBase™ on top of the Safe Imager™, and plug the short cord (a) from the Safe Imager™ into the power inlet of the iBase™ (b). Plug the connector of the power cord with the transformer into the Safe Imager™ (c), and connect the other end of the power cord to an electrical outlet.

3. Remove the gel from the package and gently remove the comb from the E-Gel® EX cassette.

4. Insert the gel into the E-Gel® iBase™ Power System, starting from the right edge. Press firmly at the top and bottom to seat the gel in the base. A steady light illuminates on the iBase™ if the cassette is correctly inserted.

5. Load gel without pre-running as follows:
   - 20 μl sample into each well
   - 20 μl appropriately diluted DNA ladder
   - 20 μl deionized water into any empty wells

   **Note:** Using DNA ladders with EDTA concentrations >0.25 mM result in low resolution and limited separation. For best results, the following DNA ladders from Invitrogen are recommended:

<table>
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<td>1% and 2% E-Gel® EX gels</td>
<td>E-Gel® 1kb Plus DNA Ladder</td>
<td>14880-090</td>
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<td>4% E-Gel® EX gels</td>
<td>E-Gel® 25bp DNA Ladder</td>
<td>14880-095</td>
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<td>E-Gel® 50bp DNA Ladder</td>
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**Run Conditions**

**Important:** Do not pre-run E-Gel® EX gels.

1. Place the amber filter over the E-Gel® iBase™.

2. Select the program, and set the run time on the iBase™ according to the percentage of the gel being run:
   - “E-Gel® EX 1−2%” (program 7) 10 minutes
   - “E-Gel® EX 4%” (program 8) 15 minutes

3. Press the Go button on the iBase™. The red light turns to a green light, indicating the start of the run.

4. The run stops automatically after the programmed time has elapsed. The end of the run is signaled by a flashing red light and rapid beeping. The LCD displays “Run Complete Press Go”.

**Opening the E-Gel® EX Cassette**

1. Place the cassette on a bench with the wells facing up.

2. Insert the sharp edge of the gel knife in the groove between the cassette halves, and lever the knife up and down. Repeat for every edge of the cassette.

3. Open the cassette and excise the band.

4. Dispose of the used gel as hazardous waste.

**Visualization and Imaging**

- View E-Gel® EX gels with an E-Gel® Safe Imager™ or other blue light transilluminator
- Always use an amber filter, or amber viewing goggles
- For imaging with a laser based scanner, verify the system has an excitation source compatible with the proprietary dye
- E-Gel® EX gels can be viewed by UV illumination, but sensitivity will be reduced
- To photograph gels with a CCD camera, a photographic filter is required e.g. SYBR Safe® filter (Invitrogen Catalog no. S37100) or Molecular Probes SYPRO® filter (Invitrogen Catalog no. S6656)
- Refer to the E-Gel® Technical Guide to determine the optimal filter sets to use, or contact the instrument manufacturer for advice
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