

Procedures for Gel Preparation with SequaGel XR[®]

SequaGel XR (EC-842) is a convenient two bottle gel system. SequaGel XR Monomer Solution contains urea as well as acrylamide and acrylamide derivatives (in a proprietary ratio) dissolved in deionized, distilled water. SequaGel Complete Buffer Solution contains 5X TBE and TEMED in deionized, distilled water. Store solutions tightly capped in a dark area at room temperature (20°C). If SequaGel Monomer Solution is refrigerated, urea may precipitate. This urea will redissolve when the solution returns to room temperature. *Acrylamide has been found to be neurotoxic. Protective gloves should be worn while handling these products. If accidental exposure occurs, contact a physician immediately.*

Mix SequaGel XR Monomer Solution and Buffer

Add appropriate volumes of SequaGel XR Monomer Solution and UreaGel Complete Buffer to a thick-walled Erlenmeyer flask (see Table 1). If desired, the solution may be degassed by stirring under vacuum for two minutes. Bring to room temperature before polymerization.

Table 1: Volumes of SequaGel XR Monomer Solution and SequaGel Complete Buffer to prepare 100mL gel solution

| SequaGel Monomer Solution | SequaGel Complete Buffer | 10% Ammonium Persulfate |
|---------------------------|--------------------------|-------------------------|
| 80 mL | 20 mL | 800 µL |

Add APS and Cast Gel

Add FRESHLY PREPARED 10% ammonium persulfate (see Table 1). Swirl gently to mix, and cast the gel. Insert the comb and allow to polymerize one to two hours. NOTE: After two hours of polymerization wrap each end of the

gel cassette with clear plastic wrap. This is important to keep the ends of the gel from drying and to maintain sample well integrity. Appropriately wrapped gels may be stored for up to 48 hours.

Suggestions for Best Results

- Clean glass plates thoroughly. Rinse with ethanol and wipe dry. Apply Glass Free (Cat.# EC-621) to one plate to ensure release after electrophoresis.
- Prerun the gel for 15-30 minutes before loading the samples. The gel temperature should be between 45-50°C.
- After the completion of the run, allow the plates to cool 10-15 minutes before separation.

Table 2: Tracking Dye Migration in SequaGel XR

| Bromophenol Blue (nucleotides) | Xylene Cyanole (nucleotides) |
|--------------------------------|------------------------------|
| 50 | 210 |

Use the table above to monitor electrophoresis progress by means of the co-migration of the bromophenol blue and xylene cyanole tracking dyes with your samples. When doing multiple loads, the next load should be added when the bromophenol blue is 3-4 cm from the bottom of the gel.

Available from National Diagnostics for Nucleic Acid Separation and Analysis

10X TBE

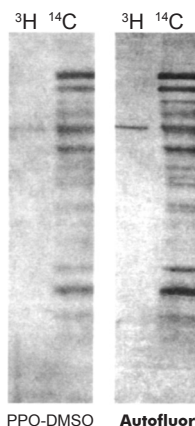
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National Diagnostics' is the purest and most stable on the market. 10X TBE contains 0.89M Tris Borate pH 8.3 and 20mM Na₂EDTA.



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