QUICK REFERENCE

invitrogen™
by life technologies™

DNA Retardation Gels

Pub. Part No. IM-6007 MAN0005886 Rev. Date 20 December 2011

Instructions are provided below for electrophoresis of DNA Retardation Gels using the XCell *SureLock*® Mini-Cell. For details, refer to the *Novex*® *Technical Guide* available at www.lifetechnologies.com/manuals or contact Technical Support.

Prepare Samples

Reagent	Sample
Sample	x µL
TBE Hi-Density Sample Buffer (5X)	1 µL
Deionized Water	to 9 µL
Total Volume	10 µL

Note

Specific buffer conditions may be required during incubation of the DNA and protein target sequence to minimize non-specific DNA/protein interactions for some samples. If the salt concentration is <0.1 M, load samples in the incubation buffer after adding 3–5% glycerol and a small amount of bromophenol blue tracking dye.

Prepare 1X Buffer

Add 100 mL 5X TBE Running Buffer to 900 mL deionized water to prepare 0.5X TBE Running Buffer.

Load Sample

Load the appropriate concentration and volume of your DNA sample on the gel.

Load Buffer

Fill the Upper Buffer Chamber with 200 mL and the Lower Buffer Chamber with 600 mL of 0.5X TBE Running Buffer.

life technologies™

DNA Retardation Gels

Run Conditions

Voltage:	100 V constant
Run Time:	90 minutes
Expected Current:	12–15 mA/gel (start); 6–15 mA/gel (end)

Blotting Conditions

For blotting DNA Retardation gels, use 0.5X TBE Running Buffer.

Perform transfer with nylon membranes at 30 V constant for 1 hour using the XCell II™ Blot Module. The expected start current is 360 mA and end current is 270 mA.

Detection

Detection is performed with ethidium bromide staining of DNA or radiolabeling the DNA or protein, for greater sensitivity.