Lonza Rockland, Inc.
www.lonza.com
biotechserv@lonza.com
Tech Service: 800-521-0390
Customer Service: 800-341-1574
Document # 18694-0307-4
Rockland, ME 04841 USA

Long Ranger® Singel® Packs

For Automated and Manual DNA Sequencing

Long Ranger Singel Packs

Long Ranger Singel Packs are designed to reduce gel preparation time and provide run-to-run consistency, while offering long, high accuracy reads. Researchers typically observe a 25%-30% increase in read length over standard polyacrylamide and an increase in base calling accuracy on automated sequencers. Long Ranger Singel Packs are compatible with all dye chemistries and are optimized for use on the ABI Prism® 377 and 373, Li-Cor® and ALFexpress® Automated DNA Sequencers.

Long Ranger Singel Packs are safe to handle. All components necessary for casting a single gel are conveniently premeasured and packaged in a ready-to-use pouch. Each component is quality assured to ensure reproducibility and consistency from run-to-run.

Long Ranger® Gel Solution-
Long Ranger Gel Solution is the core of our sequencing and genotyping product line. Due to its patented gel formulation, Long Ranger® Gels produce longer, more accurate reads.

AccuGENE® TBE Buffer- Prepared from the same qualified reagents used to produce our Gel Solutions, liquid concentrates are quality tested to ensure lot-to-lot consistency.

Precautions

Handling and use of Long Ranger Singel Packs are identical to conventional acrylamide solutions. Wear gloves and use all safety precautions required for handling acrylamide solutions. Please read the material safety data sheets (MSDS) prior to use. These products are for Research Use Only and not intended for diagnostic or human use.

Storage Conditions

Long Ranger Singel Packs should be stored at room temperature away from direct sunlight. Singel® Packs should be used before the expiration date on the packaging.

ABI Automated DNA Sequencers

	377-36 cm Sequencing GeneScan	373-34 cm Sequencing GeneScan	377-48 cm	373-48 cm
Catalog Number	50691	50693	50686	50692
Total Volume	50 ml	75 ml	50 ml	75 ml
Urea	6 M	7 M	6 M	8.3 M
Long Ranger	5%	5.75%	4.75%	5%
TBE	1X	1X	1X	1X
APS	0.05%	0.05%	0.05%	0.05%
TEMED	0.07%	0.07%	0.07%	0.07%

Manual and other DNA Sequencers

	LI-COR 0.25 mm thick gels 33 or 41 cm	AP Biotech ALFexpress	Manual
Catalog Number	50689	50685	50694
Total Volume	50 ml	75 ml	50 ml
Urea	7 M	7 M	7 M
Long Ranger®	6%	5.75%	5.75%
TBE	1.2X	1X	1X
APS	0.067%	0.05%	0.05%
TEMED	0.067%	0.07%	0.07%

AccuGENE® 5X and 10X TBE Buffers Liquid Concentrate for Automated DNA Sequencing

Prepared from qualified reagents which are used in the Long Ranger® Product Line, the AccuGENE 5X and 10X TBE Liquid Concentrates are quality tested to ensure lot-to-lot consistency.

Specifications

Components	5X	10X
	0.445 M Tris Base	0.89 M Tris Base
	0.445 M Boric Acid	0.89 M Boric Acid
	0.01 M $Na_2EDTA•2H_2O$	0.02 M $Na_2EDTA•2H_2O$
	18 MΩ water	18 MΩ water
Filtered	2 µm	2 µm
Conductivity	3.14-3.50 mS/cm	3.80-5.06 mS/cm
pH	8.25-8.38	8.0-8.5
Storage*	18°C-24°C	18°C-24°C

*TBE buffers are stable for up to 3 months from date of manufacture when stored out of direct sunlight.

For Best Results using Long Ranger® Singel® Packs

- Store Long Ranger Singel Packs at room temperature away from direct sunlight.
- Follow the recommended mixing times for the Long Ranger Singel Packs. Under-mixing will result in inadequate gel polymerization and over-mixing will result in non-polymerization.
- Ensure the glass plates are thoroughly clean and dust free. Glass plates can accumulate fluorescent contaminants from many sources including detergents, marker pens, ethanol and other solvents, poor quality water, and hands or gloves. These invisible contaminants can seriously affect data collection.
- Once the gel is polymerized (approximately 30 minutes), place paper towels soaked in electrophoresis buffer over the ends of the plates and then cover with plastic wrap. This will prevent moisture loss as the polymerization process continues. Wrapped gels may be stored at 4°C overnight.
- New matrix files may need to be prepared for ABI machines when changing reagents or dye sets.

Protocol for Long Ranger Singel Packs

Gel Preparation

1. Assemble glass plates and spacers in the cassette following the method described in your automated sequencer's manual.
2. Use the Long Ranger Singel Pack appropriate for your sequencer and plate length (See Table on page 1).
3. Remove the BLACK clip and mix the contents of the compartments by hand thoroughly, but gently, for 1 minute.
4. Place the pack on an orbital shaker for 5 minutes at medium speed.
5. Mix by hand thoroughly but gently for 1 minute.
6. Place the pack on an orbital shaker for 5 minutes at medium speed.
7. Proceed to Gel Casting.

NOTE: Do not overmix. This may interfere with gel polymerization.

Gel Casting

The following steps must be completed without delay.

1. Remove only the RED clip and mix the contents of the compartments well by hand for 1 minute.
2. Remove the WHITE clip to expose the filter to gel solution.
3. Hold the pack so the contents drain into the filter end. Fold the pack in half at the indicated line.
4. Hold the pack with the filter at the top and cut the pouch on the embossed line across the filter port.
5. Avoid introducing air into solution after mixing. Cast gel and insert comb according to your standard procedure.
6. Once the gel is polymerized (approximately 30 minutes), place paper towels soaked in electrophoresis buffer over the ends of the plates and then cover with plastic wrap. This will prevent moisture loss as the polymerization process continues.
7. Allow 2 hours for complete gel polymerization.

NOTE: Empty Long Ranger Singel Packs can be disposed of in regular trash.

Electrophoresis and Fragment Analysis with ABI Automated DNA Sequencers

Preparing for Electrophoresis

1. Remove the comb and wash the plates as described in the ABI Automated Sequencer Manual.
2. Prepare a sufficient quantity of electrophoresis buffer to fill both anodal and cathodal chambers by diluting 10X TBE stock with deionized water to 1X.
3. Mount the gel cassette onto the sequencing apparatus according to the manufacturer's instructions.
4. To assure plates and gel are clean, perform the Plate Check module specific to the dye set you are using.

NOTE: Prepare an analysis matrix standard file for Long Ranger® Gel Solution as described in the appropriate ABI Prism® Automated Sequencer Manual (377 or 373A).

ABI Prism® 377 with 36 and 48 cm Plates

1. Prepare sample sheet as normal.
2. Pre-run the gel using the desired module until a temperature of 51°C is achieved, approximately 10-20 minutes. Do not pre-run longer than necessary to reach 51°C.
3. Prepare DNA samples as you would for standard automated sequencing gels.
4. After pre-run is complete, pause the instrument and rinse the wells thoroughly with electrophoresis buffer.
5. Load samples as described in the ABI Automated Sequencer Manual.
6. Run the gel using the desired module. Run time will be 2 hours. In some cases, a 2.5 hour run time may be necessary to visualize all fragments.
7. Analyze data as usual.

ABI 373A with 48 cm Plates

1. Set the run parameters to 48-cm run length, full scan mode, 4.0% gel type, 31W power and 16-18h collection time. Set the parameters for the data collection software as usual.
2. Pre-run the gel for 10-20 minutes, then press "abort run" to stop the pre-run. Rinse the wells thoroughly with electrophoresis buffer.
3. Load samples as described in the ABI Automated Sequencer Manual.
4. Immediately after loading the last sample, press "resume run" and start data collection as usual.
5. Analyze data as usual.

ABI 373A with 34 cm Plates

1. Set the run parameters to 34-cm run length, full scan mode, 4.75% gel type, 30W power and 16h collection time. Set the parameters for the data collection software as usual. Follow steps 2-5 above.

Electrophoresis with LI-COR® Automated Sequencing Systems

Preparing for Electrophoresis

1. Prepare a sufficient quantity of running buffer to fill anodal and cathodal chambers by diluting 10X TBE stock to 1X with distilled water.
2. Remove the comb, and rinse the wells or top surface of the gel with electrophoresis buffer.
3. Mount the cassette into the sequencing apparatus according to the manufacturer's instructions.

Electrophoresis Conditions

Parameter	33 cm Standard Protocol	41 cm Standard Protocol
Data Collection Config file	33-cm-STD.col	41-cm-STD.col
Quick SequencIR Config file	Quik33cm.col	Quik41cm.col
Voltage	1500 volts	1500 volts
Current	35.0 mA	35.0 mA
Power	45.0 watts	31.5 watts
Temperature	50°C	50°C
Scan Speed	3 (=2.4 frames/hr)	3 (=2.4 frames/hr)
Pre-run time	20-30 minutes	30 minutes
Frames to Collect	25	25

Electrophoresis with the Pharmacia ALFexpress® DNA Sequencing System

Preparing for Electrophoresis

1. Prepare a sufficient quantity of running buffer to fill both anodal and cathodal chambers by diluting 10X TBE stock to 0.5X.
2. Remove the comb and rinse the wells or top surface of the gel with electrophoresis buffer.
3. Mount the gel cassette onto the sequencing apparatus according to the manufacturer's instructions.

Electrophoresis Conditions

	ALFexpress
Parameter	**Value**
Voltage	1500 Volts
Current	60 mA
Power	25 Watts
Temperature	55°C
Sampling Interval	2 seconds
Gel Thickness	0.3 mm and 0.5 mm
Run Time	15 hours

Tips and Troubleshooting for Automated Sequencing

Please consult your instrument instruction manual for tips concerning machine operation.

Polymerization

The rate of polymerization and properties of the gel depend on the quality and concentration of reagents used and attention to detail. Slow polymerization will cause gels to be more porous and weak. Fast polymerization will result in an increase in gel turbidity and decrease in gel elasticity.

It is important to note that polymerization continues long after visible gelation occurs. Gelation should occur within 15-20 minutes and complete polymerization will occur in approximately 2 hours.

- Do not mix more than 3 gels at once.
- Follow the mixing instructions carefully.
- Allow gels to polymerize for 2 hours prior to use.

Mixing Protocols: Timing and Temperature

Temperature has a direct effect on the rate of polymerization and properties of the gel. If the temperature is too high, polymerization will occur very rapidly and gels will be elastic. If the temperature is too low, polymerization will be slow and gels will be very porous.

- To ensure gel-to-gel consistency, pour gels at the same temperature.
- The optimal temperature for gel polymerization is 23°C-25°C.
- Polymerization is also highly dependent on timing, and short mixing time may result in a solution that is too cold for efficient initiation of polymerization. The pack should be at room temperature when the first clip is removed. Dissolution of urea is endothermic, and results in a drop in temperature of the pack during the first few minutes of mixing. In the twelve minutes allowed, the pack should return to room temperature after the powders are in solution. Extremely low ambient temperatures may slow polymerization.
- The timing of these processes is very important since the APS/TEMED reaction is short lived. A total of twelve minutes is allowed for the urea to dissolve and the package to return to room temperature. If there is a long delay between removing the pouch clips, polymerization may occur very slowly, making a poor gel after several hours, or there may be no polymerization at all.

Glass Plates

Use of perfectly clean apparatus and glassware is critical. In busy labs, it is easy to overlook details like dishwashing. In high throughput labs where each set of plates is used many times per week, frequent inspection of the plates is particularly important. The care applied to these tasks will be highly rewarded. Several aspects of fluorescent sequencing may be affected by the condition of the glass plates. Glass plates may accumulate fluorescent contaminants from detergents, marker pens, ethanol or other solvents, poor quality water, greasy hands or gloves etc. This is not visible to the eye but may seriously affect data collection[1].

- Glass with chipped edges may not fit correctly on the aligning pins in the ABI Prism® 377 Automated Instrument, or may cause problems at casting.
- Do not let used plates dry out before cleaning them. Dried gel is hard to remove, but very easy when still moist. Separate the plates and remove the gel by lifting with paper towels.
- Use 1%-2% Alconox® detergent and warm water. If Alconox is not obtainable in your region, consult your local ABI technical staff for a recommended alternative. Other cleaning agents may leave fluorescent deposits. Wear clean, rinsed, powder-free gloves and avoid scratching and scraping the plates. If tap water is used with Alconox, rinse the plates thoroughly after soaping with distilled or deionized water.
- Stand clean plates in an A-shaped Plate Drying Rack (Catalog No: 50641) to air dry, protected from splashes and dust (e.g. do not place it right next to the sink). They should look spotless when dry. Canned air such as Dust-Off® is useful for removing minor dust particles just before casting.
- Good dishwashing machines work well for some labs, but it is important not to let any gel dry onto the plates before running the machine. Check the washed plates carefully before use, and reject any plates with adhering particles or smears.
- Condition of combs and spacers is important: Do Not use damaged combs or spacers, since gel thickness or well formation may be affected. Variations in gel thickness and imperfections in the gel surface of the wells may cause band distortions that impair resolution.
- Note that the comb may be warped; the thin, flexible part may then form a slightly concave upper surface at the top of the gel. If this happens it is very difficult to insert the sharkstooth evenly because the teeth at the sides must be pushed well into the gel before the central teeth even touch the gel surface. This causes deformation of the wells at the sides. The tips of the sharkstooth on a warped comb form a slightly convex line, making the problem worse.
- Some labs find it useful to treat their glass plates with 2 M NaOH at regular intervals to remove traces of fluorescent contaminants. Overnight soaking is not recommended. One or two hours in NaOH, followed by thorough rinsing with deionized water, is effective. Be sure to use appropriate caution when handling the strong alkali.

Storage of the Gel

Polymerized gels may be stored up to 24 hours. To protect them from drying, after polymerization, cover the ends with paper towels dampened with 1X TBE buffer, and wrap with plastic wrap. In warm weather, storage overnight at 4°C is suggested.

Swelling of the Gel
- Some labs occasionally observe the gel swelling dramatically out of the top of the plates during electrophoresis. The swelling is caused by electroendosmosis, due to electrostatic charge on the glass surface. In this situation, cleaning with 2 M HCl is effective.

Electrophoresis Buffer
- TBE solutions should be free of precipitate. If a precipitate has formed, discard the buffer and prepare new buffer.
- Filter stock solutions through 0.45 µm filter and store in glass containers.
- Long Ranger® Singel® Packs are not compatible with Tris-TAPS EDTA (TTE) electrophoresis buffer. Use our Long Ranger® Gel Solution if a TTE buffer system is required.

Matrix File – ABI Automated DNA Sequencers
- When you change from one matrix to a different matrix, we recommend that you make a new matrix file (instrument file) on your first gel run. You need to make a separate file for each instrument, as the file is only valid for the instrument on which it was made.
- Load appropriate matrix standards and follow the easy instructions in your ABI Users Manual. Remember, do not run the base caller on the matrix standard lanes; the new file is made from the raw data.
- Mobility files cannot be changed by the user. The files supplied with the sequencer should be used, unless at some point in the future the instrument manufacturer creates new files for Long Ranger® Gels.

Red Rain

Red rain is caused by air bubbles occuring in areas where the gel is under heat stress. There are two places in the gel where bubbles can occur: the read area where the laser impact creates heat, and around the edge of the upper buffer tray above the front heat plate. Localized heat in both of these areas cannot be dissipated away by the front heat plate. Manipulations that introduce excessive air while casting will result in gels that are more prone to red rain. Long runs at high temperature or fast laser scan speeds can increase the chances of red rain appearing towards the end of the run due to prolonged heat stress. If modules are altered, the speed of the run and the quality of resolution may be changed. Increased voltage or temperature may increase the probability of red rain. Red rain may also be caused by foreign particles in the gel and the gel drying out.
- TBE solutions should be free of precipitate and filtered through 0.45 µm filters.
- Avoid any action that mixes the Gel Solutions with air.
- Cast gels in a way that avoids the introduction of too much air into the gel.
- Clean plates as suggested in previous Tips for Glass Plates.
- After gel polymerization, cover the ends with paper towels dampened with 1X TBE buffer, and wrap with plastic wrap.

Gel Dropout

Gel dropout is a fading signal around 140 bp - 220 bp. The plates seem to bind a long polymer which is released when the gel heats up during the run. Once released the polymer binds to the OH group of the migrating DNA and quenches the signal bind.
- The use of glassware dishwashers eliminates gel dropout. The combination of extremely high water temperature and prolonged physical agitation in a dishwasher effectively removes any residue buildup.
- Clean glass plates immediately after the electrophoresis run.
- Detergents and chemical washes can also be used[1].

Green or Blue Haze

Green haze in the back of the gel is common. It can be seen on the image window of the ABI Prism® 377. It is not usually strong enough to interfere with the other bands. It is caused by a buildup of fluorescent materials on the plates or misaligned plates.
- Soak plates for 20 minutes in 2 M NaOH.
- Clean glass plates immediately after the electrophoresis run.
- Clean plates as suggested in previous Tips for Glass Plates.

Long Ranger Singel Packs for Manual Sequencing

For Best Results
- Solutions of TBE containing visible precipitate should not be used. Borate can precipitate out of concentrated solutions of TBE. This will change the buffer capacity, ionic strength and the pH of the solution.
- Follow the recommended mixing times for the Long Ranger Singel Packs. Under-mixing will result in inadequate gel polymerization and over-mixing will result in non-polymerization.
- Do not use electrophoretic conditions that cause the gel to become warmer than 50°C.
- Treat only one glass plate with silanizing solution. Over silanization can cause bubbles during gel casting, gel swelling during electrophoresis and gel detachment from the glass plate.
- **To minimize gel swelling during fixing, a 20% ethanol and 10% acetic acid fixing solution should be used.**

Gel Preparation

1. Glass plates must be clean and free of dried gel and soap residues. To remove residues, apply ethanol to both plates and wipe dry.
2. Ensure that the gel will not stick to the glass plates by silanizing only one plate.
3. Assemble glass plates according to manufacturer's instructions.
4. Use the Long Ranger® Singel ® Pack appropriate for manual sequencing.
5. Remove the BLACK clip and mix the contents of the compartments by hand thoroughly but gently for 1 minute.
6. Place the pack on an orbital shaker for 5 minutes at medium speed.
7. Mix by hand thoroughly but gently for 1 minute.
8. Place the pack on an orbital shaker for 5 minutes at medium speed.
9. Proceed to Gel Casting.

NOTE: Do not overmix. This may interfere with gel polymerization.

Gel Casting

The following steps must be completed without delay.

1. Remove only the RED clip and mix the contents of the compartments well by hand for 1 minute
2. Remove the WHITE clip to expose the filter to gel solution.
3. Hold the pack so the contents drain into the filter end. Fold the pack in half at the indicated line.
4. Hold the pack with the filter at the top and cut the pouch on the embossed line across the filter port.
5. Avoid introducing air into solution after mixing. Cast gel and insert comb according to your standard procedure.
6. Once the gel is polymerized (approximately 30 minutes), place paper towels soaked in electrophoresis buffer over the ends of the plates and then cover with plastic wrap. This will prevent moisture loss as the polymerization process continues.
7. Allow 2 hours for complete gel polymerization.

NOTE: Empty Long Ranger Singel Packs can be disposed of in regular trash.

Preparing for Electrophoresis

1. Prepare a sufficient quantity of running buffer to fill both anodal and cathodal chambers by diluting 10X TBE stock to 0.6X or 1X with deionized water, as appropriate.
2. Remove the comb, and rinse the wells or top surface of the gel with electrophoresis buffer.
3. Mount the gel cassette onto the sequencing apparatus according to the manufacturer's instructions.

Autoradiography

1. When the run is complete, turn off the power supply, disconnect electrodes and remove the plates from the apparatus.
2. Long Ranger® Gels do not require fixing or removal of urea for ^{35}S labeled reactions. If your application requires fixing, soak in 20% ethanol and 10% acetic acid for 10 to 20 minutes.
3. Allow the plates to cool briefly before separating. Transfer the gel onto Whatman® 3MM chr filter paper.
4. Place the Whatman 3MM chr paper on a flat surface with gel side up and cover with plastic wrap.
5. Dry the gel (not required for ^{32}P-labeled gels) under vacuum at 70°C-80°C for 30-60 minutes.
6. Remove the plastic wrap and expose gel to X-ray film using standard techniques.

Electrophoresis

1. Pre-run the gel for 10 to 15 minutes before loading the samples. Use 28 - 35 W for 35 ml gels (40 cm x 20 cm x 0.4 mm) or 55 - 66 W for 70 ml gels (40 cm x 40 cm x 0.4 mm).
2. Prepare DNA samples as you would for standard sequencing gels: denature samples for 2 to 5 minutes at 75°C or the temperature recommended for the enzyme you are using, and immediately chill on ice.
3. After the pre-run, rinse the well(s) thoroughly with electrophoresis buffer. Reinsert the sharks-tooth comb so it just touches the gel, and load samples.
4. Adjust the power so that the temperature of the glass plates is between 40°C - 50°C. We recommend using 28 - 35 W for 35 ml gels and 55 - 66 W for 70 ml gels. Monitor the temperature of the plates, and adjust wattage to maintain the desired temperature.
5. Monitor the run time with the marker dyes, bromophenol blue and xylene cyanol.

Approximate Running Times of Long Ranger Gels			
Bases Read	**250**	**400-500**	**>600 (double load)**
Pre-run	10-15 minutes	10-15 minutes	10-15 minutes
Run Time	2 hours	4 hours	6-8 hours

Dye Migration in Long Ranger Gels (1X TBE)		
Gel %	**Bromophenol Blue**	**Xylene Cyanol**
5	40 bp	175 bp
6	20 bp	138 bp
7	19 bp	123 bp
8	11 bp	98 bp

Tips and Troubleshooting for Manual Sequencing

Polymerization

The rate of polymerization and properties of the gel depend on the quality and concentration of reagents used and attention to detail. Slow polymerization will cause gels to be more porous and weak. Fast polymerization will result in an increase in gel turbidity and decrease in gel elasticity.

It is important to note that polymerization continues long after visible gelation occurs. Gelation should occur within 15-20 minutes and complete polymerization will occur in approximately 2 hours.

- Do not mix more than 3 gels at once.
- Follow the mixing instructions carefully.
- Allow gels to polymerize for 2 hours prior to use.

Mixing Protocols: Timing and Temperature

Temperature has a direct effect on the rate of polymerization and properties of the gel. If the temperature is too high, polymerization will occur very rapidly and gels will be elastic. If the temperature is too low, polymerization will be slow and gels will be very porous.

- To ensure gel-to-gel consistency, pour gels at the same temperature.
- The optimal temperature for gel polymerization is 23°C-25°C.
- Polymerization is also highly dependent on temperature, and short mixing time may result in a solution that is too cold for efficient initiation of polymerization. The pack should be at room temperature when the first clip is removed. Dissolution of urea is endothermic, and results in a drop in temperature of the pack during the first few minutes of mixing. In the twelve minutes allowed, the pack should return to room temperature after the powders are in solution. Extremely low ambient temperatures may also slow polymerization.
- The timing of these processes is very important since the APS/TEMED reaction is short lived. A total of twelve minutes is allowed for the urea to dissolve and the package to return to room temperature. If there is a long delay between removing the pouch clips, polymerization may occur very slowly, making a poor gel after several hours, or there may be no polymerization at all.

Glass Plates

- Glass plates should be free of dried gel material. Wash plates immediately after removal of the gel.
- Plates can be washed with a 0.5% solution of Alconox®, rinsed with hot water and wiped with ethanol.
- Dirt and debris on glass plates can cause bubbles to form during gel casting. Wash plates as described in the previous bullet point.
- To avoid gel sticking, treat one glass plate with a silanizing agent (Gel Slick® Catalog No. 50640)
- Over silanization can cause gels to swell between the glass plates and may cause sample loading problems. Both plates should be stripped by soaking in a 2 M NaOH solution for 30 minutes and then re-coat one plate only with a silanizing agent.

Electrophoresis Buffer

Buffer problems can cause a host of gel problems. Some problems include fuzzy bands, localized heating, smiling, frowning, wavy bands and slanted bands. A breakdown in the buffering system can also cause problems at the interface between the gel and the running buffer. You may see swelling of the gel out of the top or bottom of the cassette and an uneven well area.

- TBE solutions should be free of precipitate. If a precipitate has formed, discard the buffer and prepare new buffer.
- The use of a discontinuous buffer system is not recommended.
- Long Ranger® Singel® Packs are not compatible with glycerol tolerant buffer (Tris-Taurine EDTA). Use our Long Ranger Gel Solution in these applications.

Electrophoresis Conditions

- Long Ranger® Gels should not exceed a temperature of 51°C during electrophoresis.
- Gels should be pre-run to a temperature of 45°C-50°C prior to loading samples.

Fixative Solution

Because of Long Ranger® Solution's unique structure, gels will not crack during drying even if they are not fixed.

- If your protocol requires fixing, use a solution of 20% ethanol and 10% acetic acid to minimize gel swelling.
- Use the correct fixative as stated above to avoid the gel from swelling while in the fixative.
- Over-silanization of glass plates can cause the gel to detach from the plate during fixing. Strip both glass plates by soaking in a 2 M NaOH solution for 30 minutes and then re-coat one plate only with a silanizing agent.

Sample Preparation

- Samples should be denatured for 2 to 5 minutes at 75°C-80°C prior to loading on the gel.
- If the sequencing reaction is narrowing near the bottom of the gel, your sample could contain excessive salt. Wash the template DNA an additional time with 70% ethanol.
- If you are using a DNA polymerase with high glycerol concentration, substitute TTE buffer for TBE buffer and use our Long Ranger Gel Solution.

References

1. Applied Biosystems (2001). Cleaning Glass Plates to eliminate temporary loss of signal. P/N 4306162
2. Applied Biosystems (2001). Achieving longer High-Accuracy Reads on the 377 sequenser. P/N 4315153
3. Shorr, Robert. (1993). Electrophoretic Media. U.S. Patent 5219923.
4. Lonza, Inc (1998). Long Ranger® Singel® Packs for automated sequencing [ReSOURCE Notes] #01-0298.

Ordering Info

Part Number	Description	Size Quantity
50691	Long Ranger Singel Pack - ABI Prism® 377-36 cm	5 pack
50686	Long Ranger Singel Pack - ABI Prism 377-48 cm	5 pack
50693	Long Ranger Singel Pack - ABI Prism 373-34 cm	5 pack
50692	Long Ranger Singel Pack - ABI Prism 373 -48 cm	5 pack
50689	Long Ranger Singel Pack – LI-COR® 33 or 41 cm	5 pack
50685	Long Ranger Singel Pack - ALFexpress®	5 pack
50694	Long Ranger Singel Pack - Manual Sequencing	5 pack
50839	AccuGENE® 5X TBE Buffer	4 L
50835	AccuGENE 5X TBE Buffer	10 L
50836	AccuGENE 5X TBE Buffer	20 L
50843	AccuGENE® 10X TBE Buffer	1 L
50840	AccuGENE 10X TBE Buffer	4 L
50837	AccuGENE 10X TBE Buffer	10 L
50838	AccuGENE 10X TBE Buffer	20 L

Related Products

Part Number	Description	Size Quantity
50611	Long Ranger® 50% Gel Solution	250 ml
50615	Long Ranger 50% Gel Solution	1 L
50640	Gel Slick® Solution	250 ml
50641	Plate Drying Rack	Each

For Research Use Only.

Long Ranger, Gel Slick, AccuGENE and Singel are trademarks of Lonza Group or its subsidiaries. Long Ranger is covered by U.S. Patent #5,219, 923.
LI-COR is a trademark of LI-COR, Inc. ALFexpress is a trademark of Pharmacia Biotech AB Corporation.
Nalgene is a trademark of Nalge Company Corp. Whatman is a trademark of Whatman Paper Limited Company.
Dust-off is a trademark of Falcon Safety Products, Inc. Alconox is a trademark of Alconox Corporation.
ABI Prism is a registered trademark of PE Corporation, Applied Biosystems Division.

© 2007 Lonza Rockland, Inc.
All Rights Reserved.