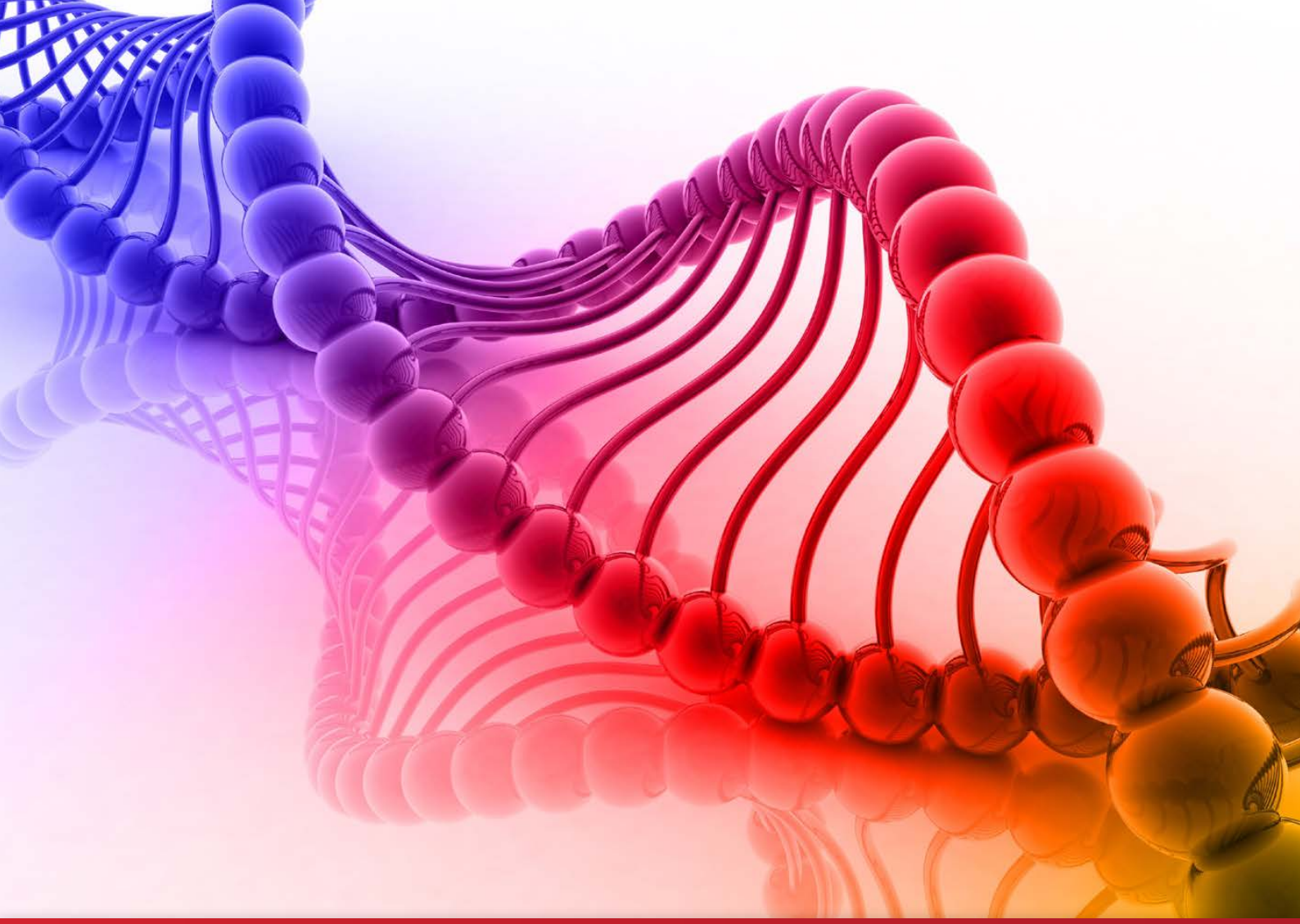


Molecular Cloning Laboratories
Sanger Sequencing Portfolio

Significant Cost Savings vs ABI
Uncompromised Performance
Longer Read Length
Longer Shelf Life
No Recalibrations Necessary

GeneTarget*Solutions*

MCLAB
Molecular Cloning Laboratories

BrightDye® Terminator Cycle Sequencing Kit

The BrightDye® Terminator Cycle Sequencing Kit is designed for de novo sequencing, resequencing, PCR product, plasmid, fosmid, and BAC templates by utilizing highly flexible chemistry. This kit uses four different fluorescent dyes to label ddNTPs, which are added sequentially to the primer through a cycle sequencing reaction. All required reagents for the sequencing procedure are in a reaction-ready, pre-mixed format. BrightDye® Terminator kit is compatible with commercially available BigDye® Terminator V3.1 kit, without any changes in the workflow.

With enhanced robustness and better dye mobility characteristics, the BrightDye® Terminator Cycle Sequencing Kit can be used in a wide range of applications with the following features:

- Long read lengths.
- Better performance reading through GC rich regions.
- Longer reads with more uniform peak heights.
- Improved productivity for less cost.

BrightDye® Terminator Kit – superior to BigDye® Terminator v3.1 Kit in sequencing pGem templates:

Figure A. Bigdye® Terminator v3.1 dilution fold 32X: 1.875 µl 5X dilution buffer (ABI) + 0.250 µl BigDye® Terminator v3.1, total reaction volume 10 µl

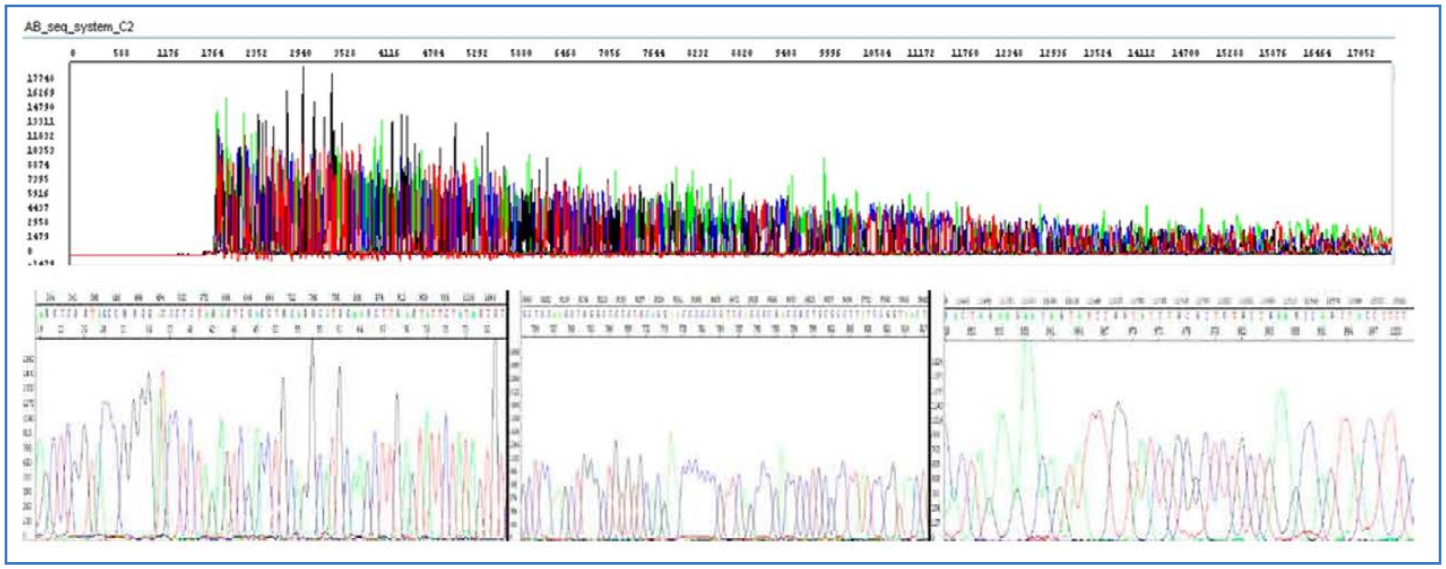
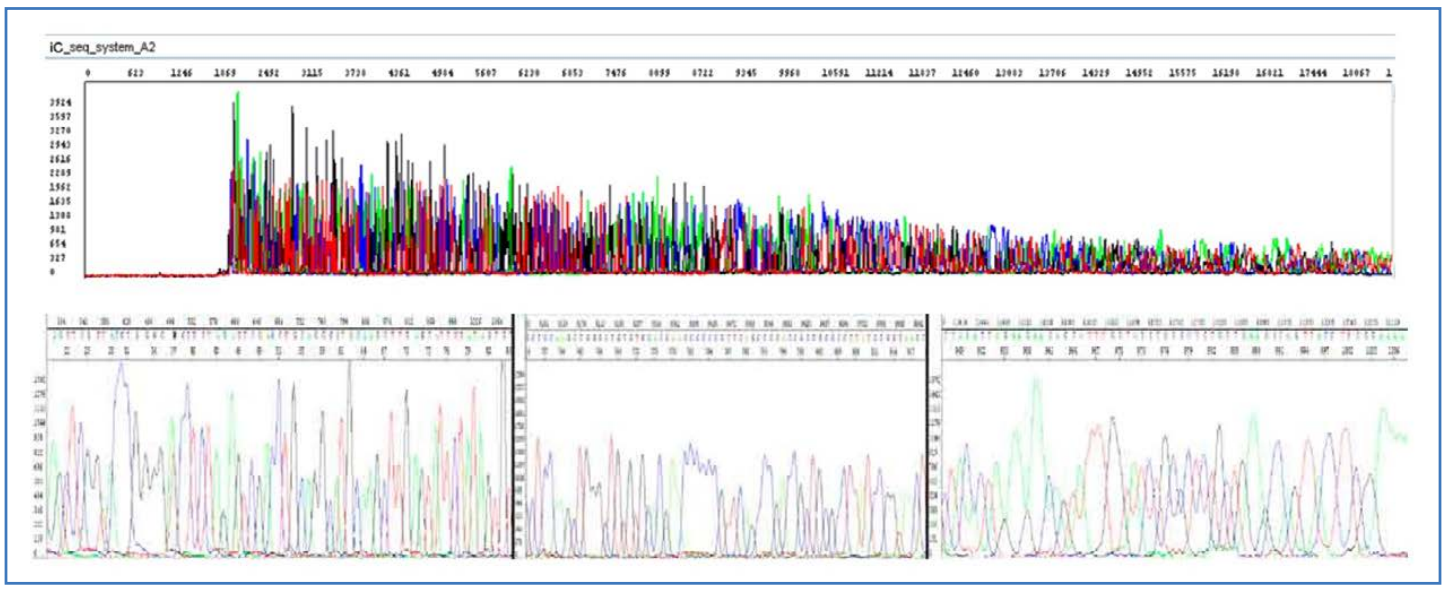


Figure B. BrightDye® Terminator (MCLAB) dilution fold 32X: 1.875 µl 5X dilution buffer (MCLAB) + 0.250 µl BrightDye® Terminator (MCLAB), total reaction volume 10 µl



BrightDye® Terminator Kit – superior to BigDye® Terminator v3.1 Kit in sequencing difficult templates:

Figure C. BigDye® Terminator v3.1 dilution fold 32X: 1.875 µl 5X dilution buffer (ABI) + 0.250 µl BigDye® Terminator v3.1, total reaction volume 10 µl

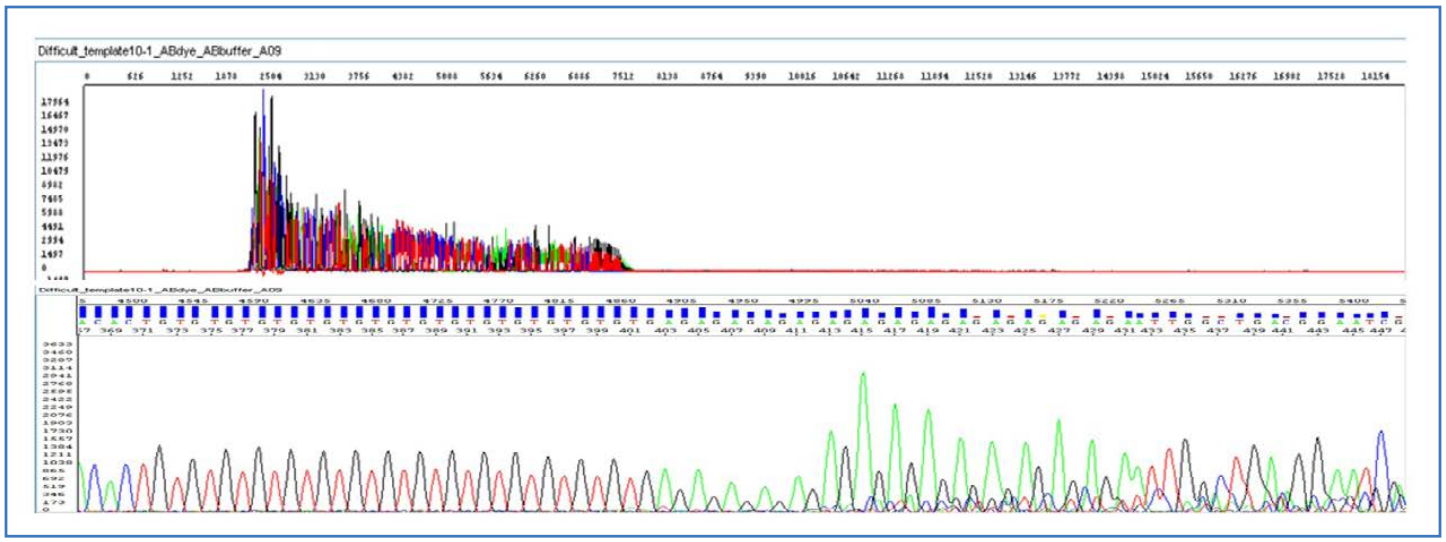
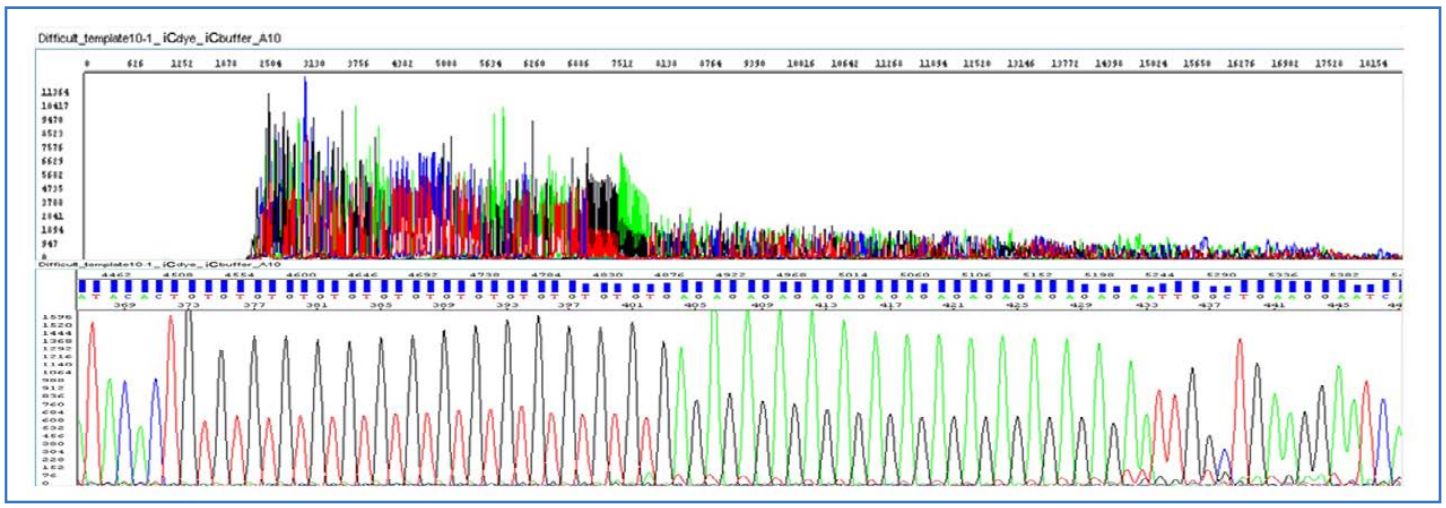


Figure D. BrightDye® Terminator (MCLAB) dilution fold 32X: 3.750 µl BDX64 buffer (MCLAB) + 0.250 µl BrightDye® Terminator (MCLAB), total reaction volume 10 µl. Peak uniformity and high quality base calls using BrightDye® Terminators.



Applications:

- De novo sequencing
- Resequencing
- Sequencing difficult templates
- Long-read sequencing
- Sequencing across all template types (plasmids, PCR products, BACs, and fosmids)
- Mixed-base detection
- Sequencing short PCR products using rapid electrophoresis run modules

Platforms:

GeneAmp® 9700, Veriti® Thermal Cycler, 3730 DNA Analyzer, 3500xL Genetic Analyzer, 3500 Genetic Analyzer, 310 Genetic Analyzer, 3130 Genetic Analyzer, 3130xl Genetic Analyzer, 3730xl DNA Analyzer

Template Compatibility:

- Plasmid DNA (≤15Kb)
- Fosmids
- Single Stranded DNA
- Lambda DNA
- BAC DNA
- Genomic DNA (Bacterial)
- PCR Amplicons

Storage:

Store kit at -20°C. Avoid repeated freeze-thaw cycles.

Cat #	Size
BDT3-24:	24 reactions
BDT3-100:	100 reactions
BDT3-200:	2,500 reactions
BDT3-300:	5,000 reactions (2 x 20ml bottles)
BDT3-400:	25,000 reactions

NanoPOP™ Polymers

NanoPOP™4, NanoPOP™6, and NanoPOP™7 are separation matrixes formulated from nanoparticles based on MCLAB's "Block Copolymer Technology" chemistry. The new matrixes have better coating and separating abilities. They are designed for ABI Genetic Analyzers with different applications. Customers can use their current run modules and protocols without any change. New spectral calibration is not needed.

Application:

NanoPOP™4: denaturing DNA fragment analysis such as microsatellite and SNP genotyping
NanoPOP™6: standard and rapid DNA sequencing
NanoPOP™7: DNA sequencing and fragment analysis

Size 5ml, for 310/3100, 3130/3130XL Genetic Analyzer Accessories.

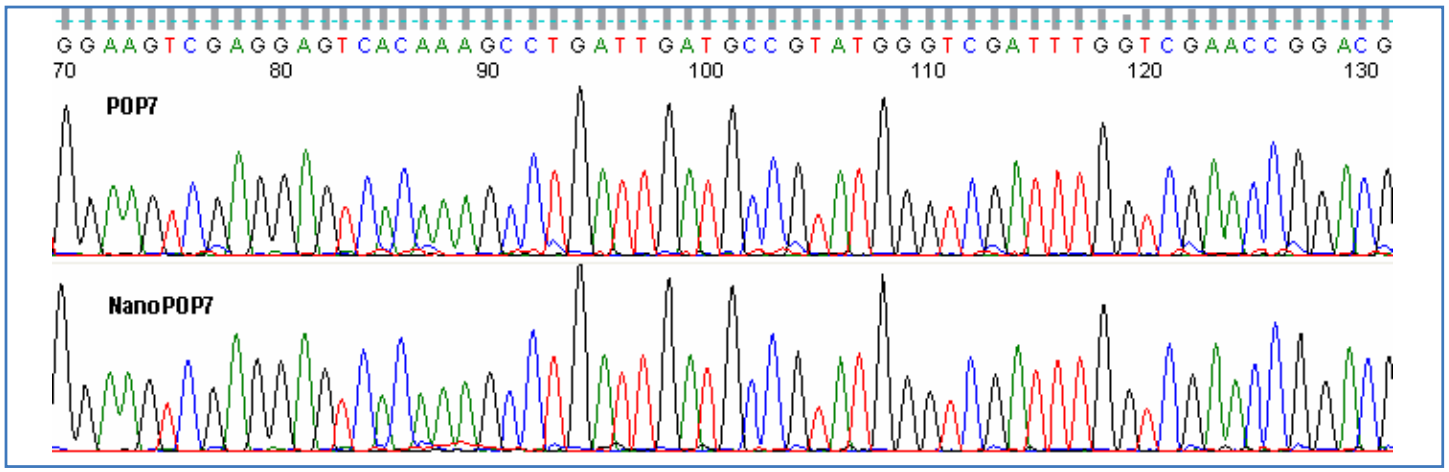
Size 10ml, for 3130/3130XL Genetic Analyzer Accessories.

Size 10ml, for 310/310v0 Genetic Analyzer Accessories.

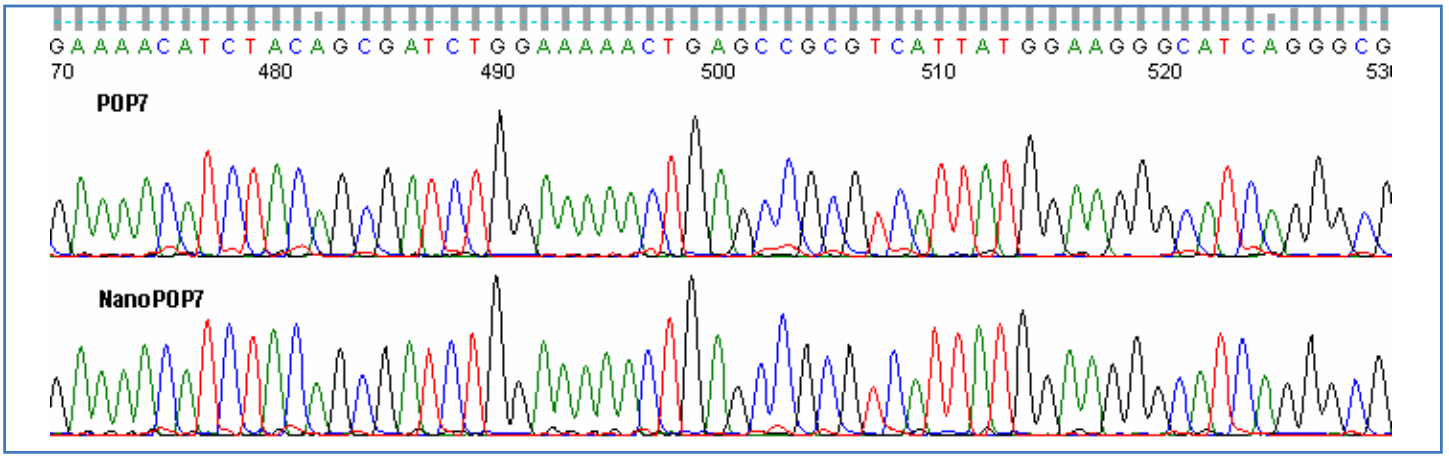
Size 28ml, for 310/3100, 3130/3130XL, 3730/3730XL Genetic Analyzer Accessories.

Comparisons:

1. NanoPOP™7 vs POP™7: near 100 bp

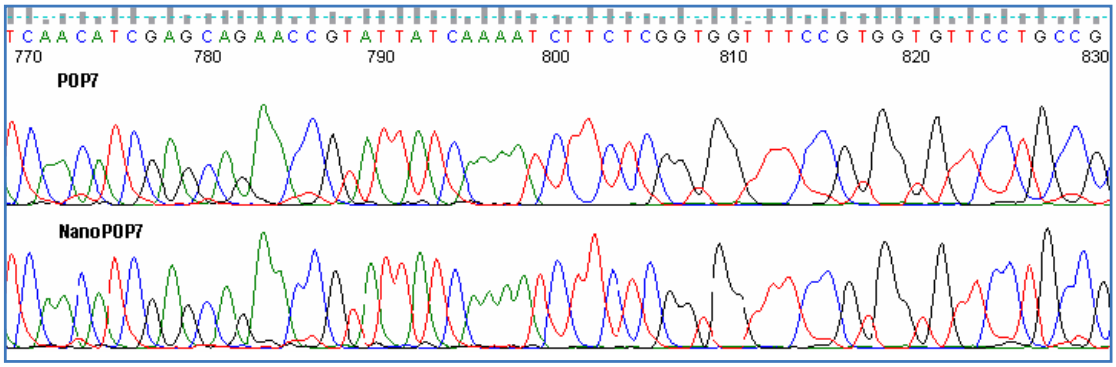


2. NanoPOP™7 vs POP™7: near 500 bp

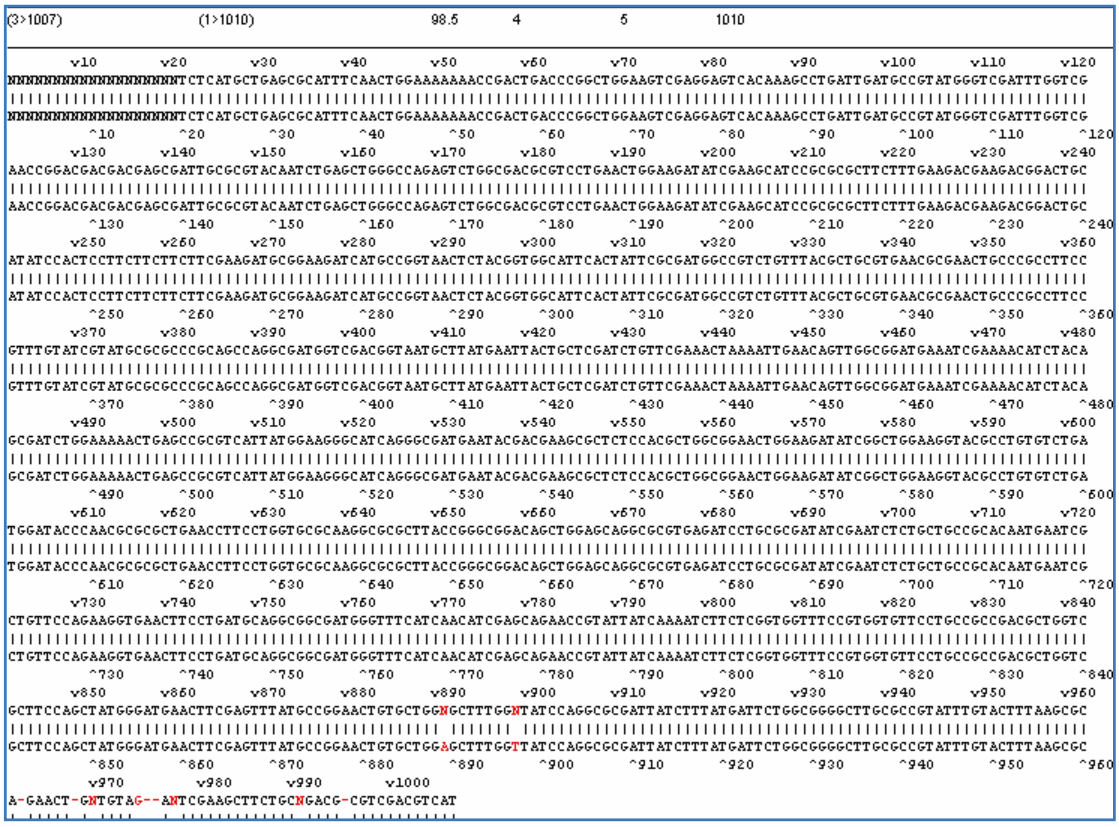


Comparisons continued:

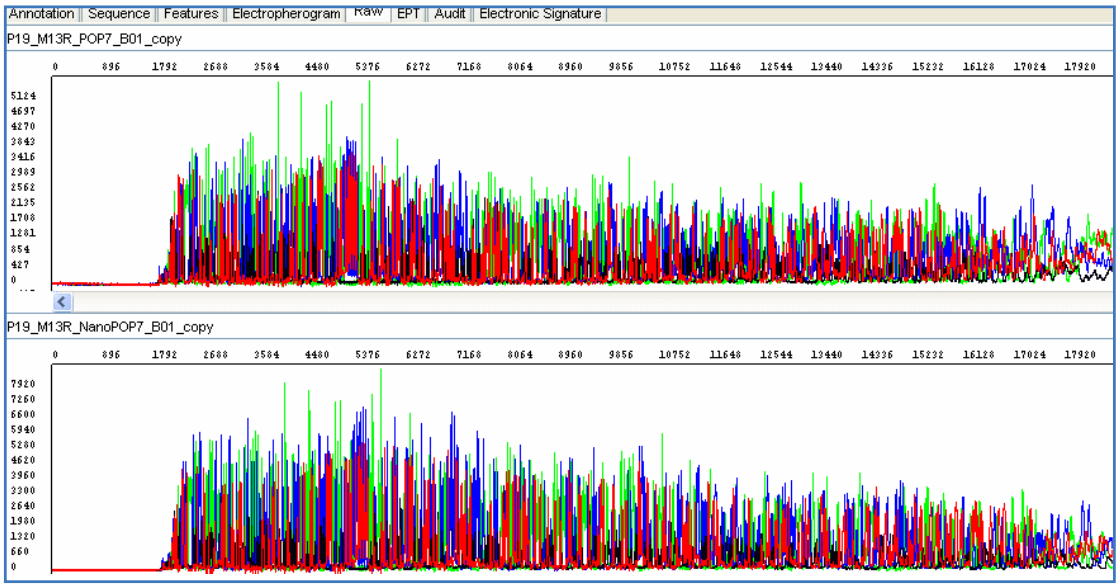
3. NanoPOP™7 vs POP™7: near 800 bp



4. Alignment



5. Raw Data



Name	Cat#	Size
NanoPOP™4	NP4-100	3130/3130xl Genetic Analyzers (ABI), 5ml
NanoPOP™4	NP4-101	3130/3130xl Genetic Analyzers (ABI), 10ml
NanoPOP™4	NP4-102	3130/3130xl Genetic Analyzers (ABI), 28ml
NanoPOP™4	NP4-120	310 Genetic Analyzers (ABI), 5ml
NanoPOP™4	NP4-121	310 Genetic Analyzers (ABI), 10ml
NanoPOP™4	NP4-122	310 Genetic Analyzers (ABI), 28ml
NanoPOP™6	NP6-100	3130/3130xl Genetic Analyzers (ABI), 5ml
NanoPOP™6	NP6-101	3130/3130xl Genetic Analyzers (ABI), 10ml
NanoPOP™6	NP6-120	310 Genetic Analyzers (ABI), 5ml
NanoPOP™6	NP6-121	310 Genetic Analyzers (ABI), 10ml
NanoPOP™7	NP7-100	3130/3130xl Genetic Analyzers (ABI), 5ml
NanoPOP™7	NP7-101	3130/3130xl Genetic Analyzers (ABI), 10ml
NanoPOP™7	NP7-300	3130/3130xl, 3730/3730xl Genetic Analyzers (ABI), 28ml
NanoPOP™7	NP7-301	3730/3730xl Genetic Analyzers (ABI), 10 x 28ml
NanoPOP™7	NP7-302	3730/3730xl Genetic Analyzers (ABI), 30 x 28ml

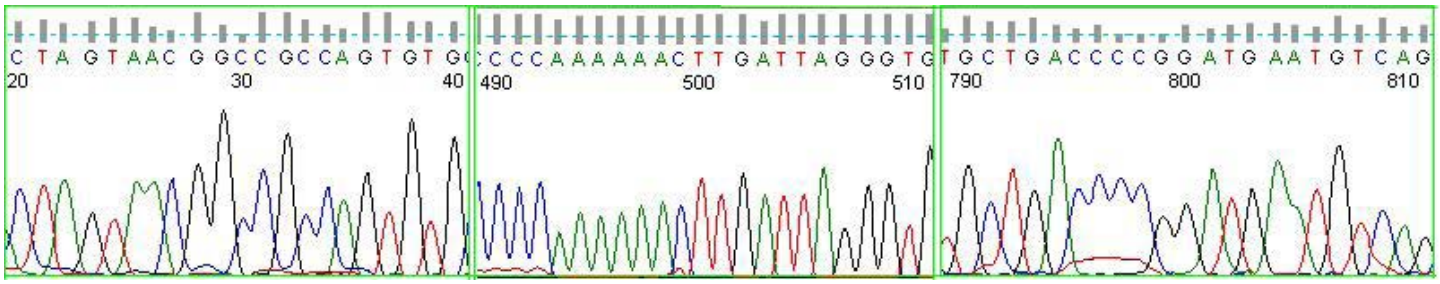
BDX64 (BigDye® Enhancing Buffer)

MCLAB's BDX64 is a BigDye® enhancing buffer. It has the same ion strength as the BigDye® premix 3.1 and 1.1. Up to 64 (0.12 µl BigDye® in 10µl reaction) or more fold dilutions by combining with the 5x dilution buffer. Enhances the polymerase activity and reduces the extension time from 4 to 1 minute. Reduces signal decline rate and results in even peak distribution. Optimizes for use with BigDye® Chemistry (ABI) on 310, 3100, 3130/3130xl & 3730/3730xl.

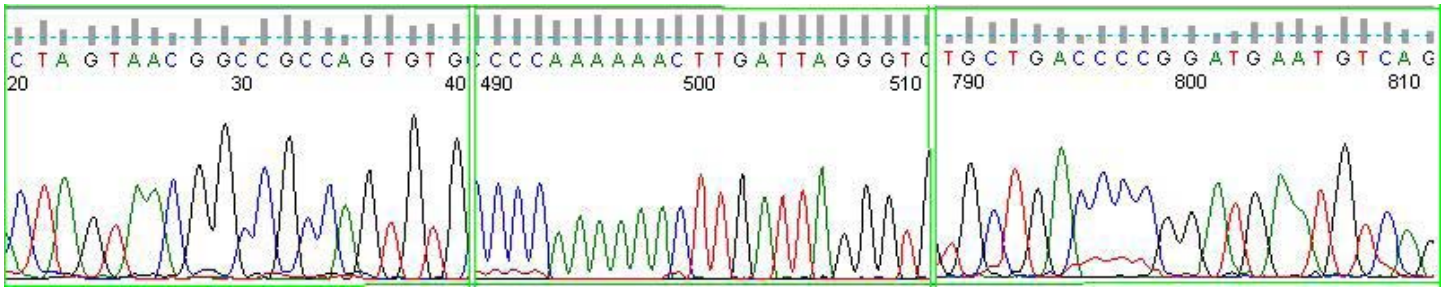
An Application Example (64 fold):
BDX64: 0.875µl
BigDye®3.1: 0.125µl
5X dilution buffer: 1.5µl (catalog # is SBUF-100)
Template: 100ng
Primer: 3.2pmol
H2O: 10µl

Sequencing Examples:

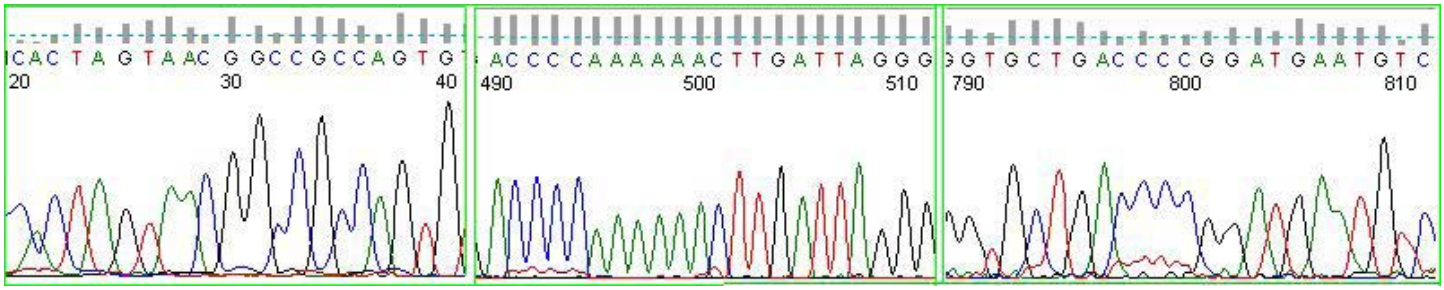
1. Dilution fold 32X:
1.5µl 5X dilution buffer + 0.750µl BDX64 buffer + 0.250µl BigDye® Terminator 3.1, total reaction volume 10µl



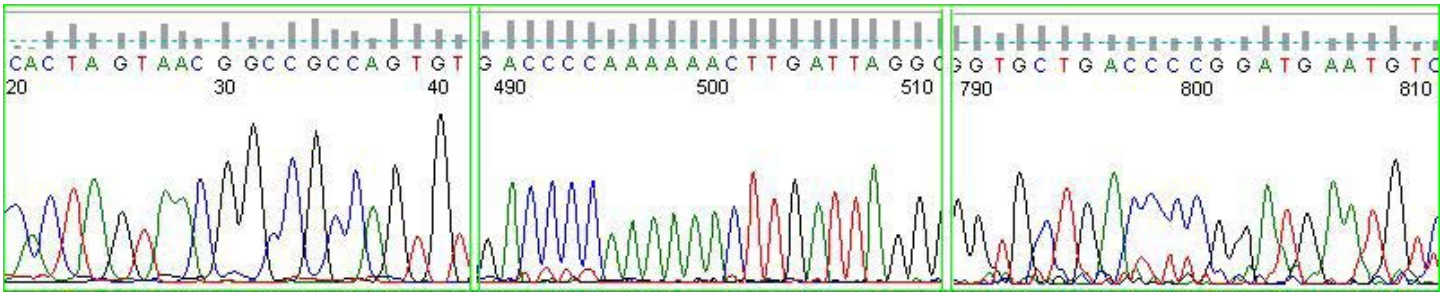
2. Dilution fold 64X:
1.5µl 5X dilution buffer + 0.875µl BDX64 buffer + 0.125µl BigDye® Terminator 3.1, total reaction volume 10µl



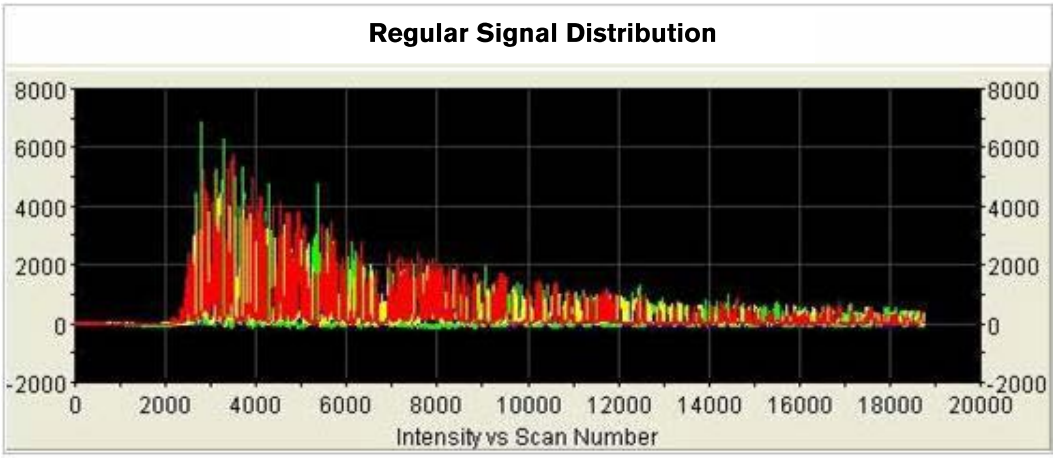
3. Dilution fold 128X:
1.5µl 5X dilution buffer + 0.937µl BDX64 buffer + 0.063µl BigDye® Terminator 3.1, total reaction volume 10µl

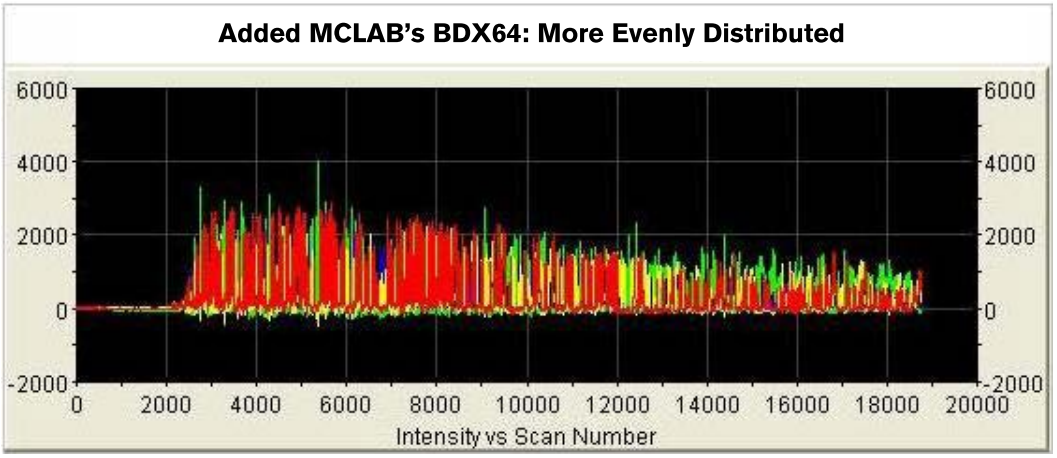


4. Dilution fold 256X:
1.5µl 5X dilution buffer + 0.969µl BDX64 buffer + 0.031µl BigDye® Terminator 3.1, total reaction volume 10µl



Signal Distributions: before and after BDX64 was added





Recommended Cycle Conditions: 96°C, 3 minutes. + 30x (96°C, 10 seconds. 50°C, 5 seconds. 60°C, 2 mins.)

Recommended Storage Conditions: -20°C

Name	Cat #	Size
BDX64 Buffer	BDX-100	2 x1.25 ml

BigDye® Sequencing Clean Up Kit

BigDye® Sequencing Clean Up Kit is a magnetic bead based high-throughput purification of DNA sequencing reaction kits. The kit consists of beads and elution buffer. Each component has been optimized for removing salts and unincorporated dye terminators from DNA sequencing reaction mixtures. The purified DNA products are more stable compared to products purified by using competitors' magnetic beads. The system can be easily adapted in your current system. You may use the same protocol as using CleanSeq® beads.

Recommended Storage Conditions: 4°C

Name	Cat #	Size
BigDye® Cleaning Beads	BCB-100	5 mL
BigDye® Cleaning Beads	BCB-200	50 mL
BigDye® Cleaning Beads	BCB-300	500 mL

BigDye® Terminator 5X Sequencing Buffer

The BigDye® Terminator 5X Sequencing Buffer reduces sequencing costs without affecting sequence accuracy or read length. Its buffer system enables to use less dye terminator mix.

Application: DNA sequencing

Recommended Storage Conditions: 4°C

Recommended Protocol:

Prepare sequencing reactions (10µl rxn) according to the following:
- Add dye terminator mix 0.1-0.2µl
- Add 5X Sequencing Buffer 2µl
- Add template (100-500 ng/µl) 1 µl
- Add primer (3 pmol/µl) 1 µl
- Add H2O to 10 µl

Cycle the reaction according to the following protocol: 30 seconds, 96°C => 15 seconds, 50°C => 4 minutes, 60°C => Cycle 25 times.

Notes: Other cycle conditions may work well for individual users. These conditions work well for most samples processed in our labs using reduced volume and reduced Terminator mix concentrations.

Name	Cat #	Size
BigDye® Terminator 5X Sequencing Buffer	SBUF-100	1 mL
BigDye® Terminator 5X Sequencing Buffer	SBUF-110	28 mL
BigDye® Terminator 5X Sequencing Buffer	SBUF-120	233 mL

CARE Solution

MCLAB's CARE solution is designed for inline capillary regeneration. It has been tested on ABI's 310, 3100, 3130xl and 3730xl successfully. Besides capillary arrays, the CARE solution can also clean polymer contacted surfaces (e.g. Pump channels, Polymer block and tubings). The inline capillary regeneration protocol makes system cleaning easier than ever and the regenerated capillary has lower background noise, and longer sequencing reads.

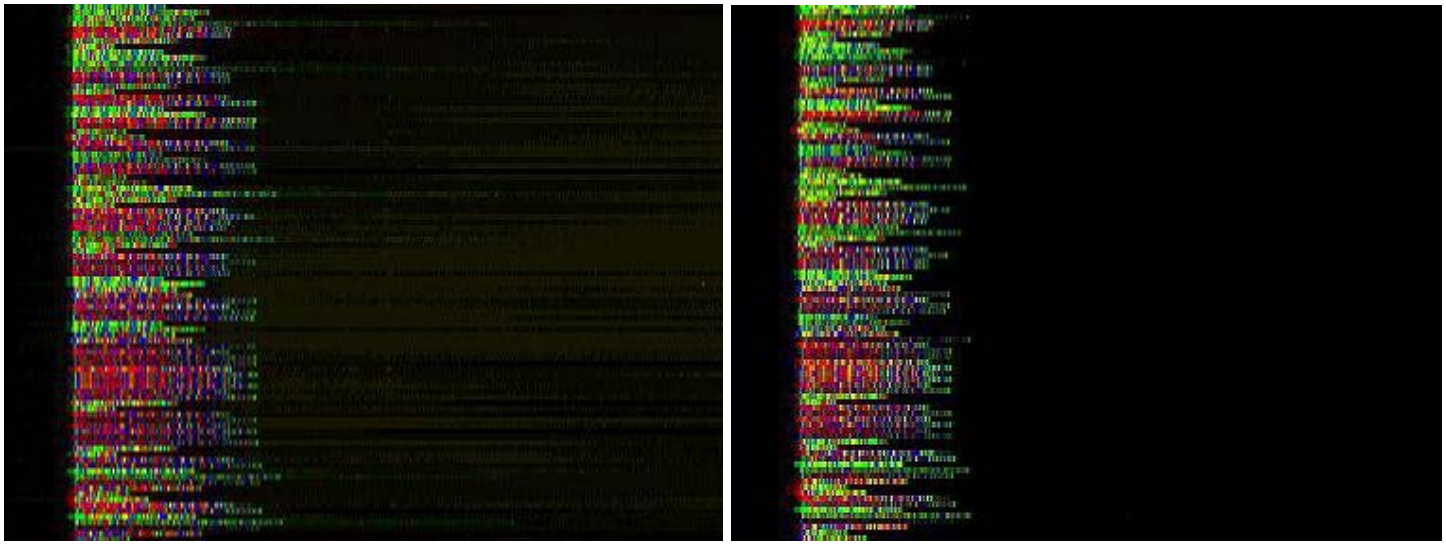
Application:

- Capillary inline rejuvenation
- Pump channels, Polymer block and tubing cleaning
- Yellow haze background removal

Recommended Storage Conditions: 4°C.

Comparisons:

1. Before CARE applied (left) vs. After CARE applied (right)



Name	Cat #	Size
CARE Solution	CR-100	28 ml
CARE Solution	CR-500	5 x 28 ml

DNA Size Standard

MCLAB's DNA Size Standard series products are internal lane standards that are intended to be used in assigning sizes to DNA fragments on fluorescence-detecting instruments. Common applications include genotyping and DNA Fragment Analysis. Each of these standards consists of 15 DNA fragments, ranging in 50, 75, 100, 139, 150, 160, 200, 250, 300, 340, 350, 400, 450, 490 and 500 bp. Each band is single-stranded and fluorescence-labeled either with carboxyx-rhodamine (ROX dye) or MCLAB's proprietary fifth orange dye. Size fragments are evenly distributed and can be used for very accurate size calling.

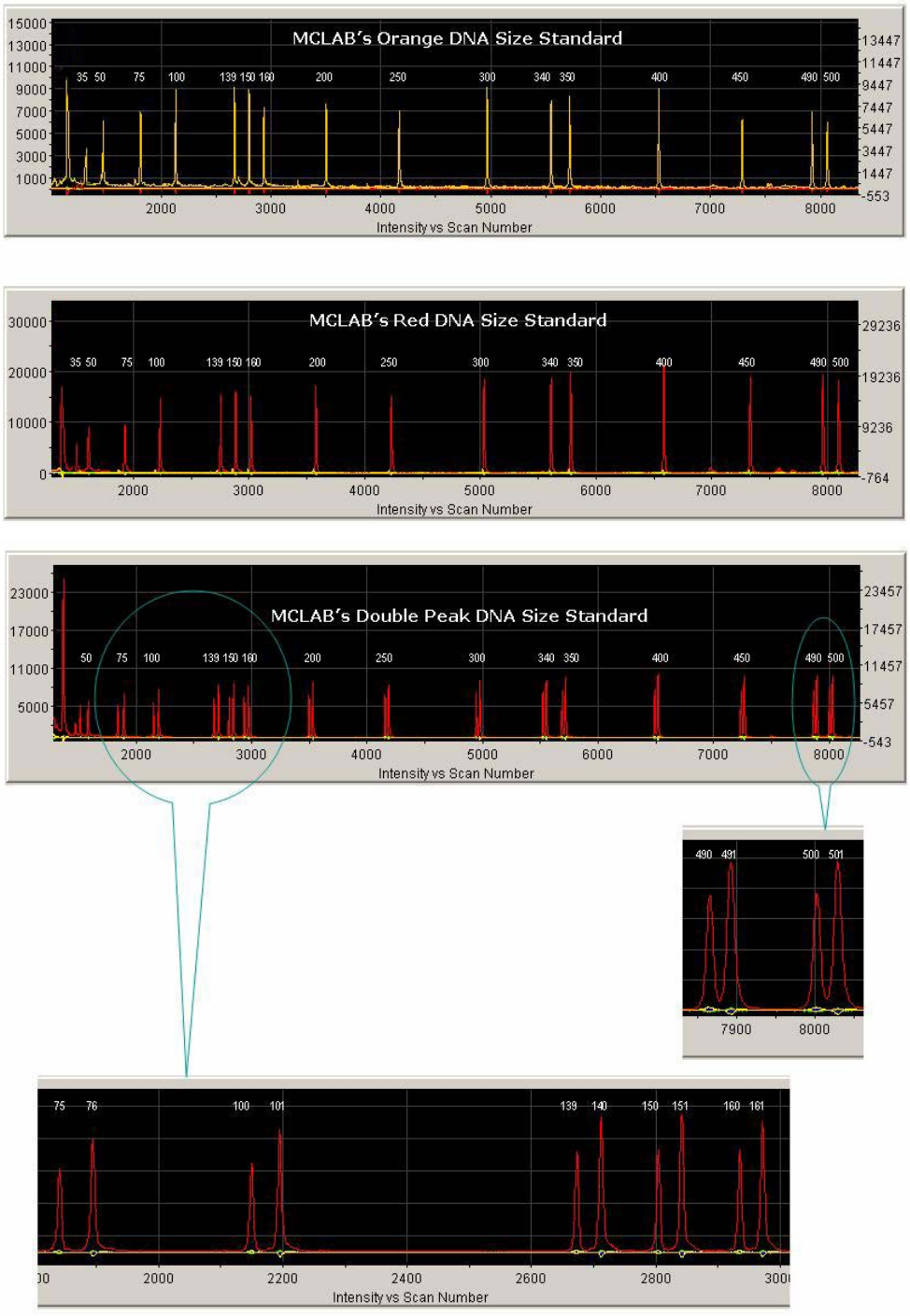
Red DNA Size Standard: has been adapted on DNA fragment analysis software, e.g. GeneMapper™ (ABI) and GeneScan™ (ABI) by using same parameters as ABI's GeneScan™ 500 ROX™ Size Standard. **Orange DNA Size Standard:** can be used at the same setting as ABI's GeneScan 500 Liz Size Standard. **Double Peak DNA Size Standard:** similar to Red/Orange DNA Size Standard, except every fragment is accompanied by an one-base-plus band. For example, 50 becomes 50 and 51, 75 become 75 and 76, etc. This single base separation is very useful for monitoring the performance of the installed capillary arrays.

Product Formats:
(1) Normal (DSMR-100, DSMO-100, DSMD-100)
(2) Premixed in Super-DITM Formamide (DSMR-101, DSMO101, DSMD-101)

Recommended Loading:
(1) Normal: 0.5µl per well
(2) Premixed in Super-DI™ Formamide: 15µl per well
The premixed size standard products are ready-to-use and can be aliquoted into the plate well directly.

Recommended Storage Conditions: Stable at 4°C or -20°C for 6 months Avoid repeatedly freeze-thawing.

Examples:



Name	Cat #	Size
Red DNA Size Standard	DSMR-100	800 analyses (400µl)
Red DNA Size Standard	DSMR-101	800 analyses (8 x 1.5ml, premixed in Super-DI™)
Orange DNA Size Standard	DSMO-100	800 analyses (400µl)
Orange DNA Size Standard	DSMO-101	800 analyses (8 x 1.5ml, premixed in Super-DI™)
Double Peak DNA Size Standard	DSMD-100	800 analyses (400µl)
Double Peak DNA Size Standard	DSMD-101	800 analyses (8 x 1.5ml, premixed in Super-DI™)

Genotyping Reference Human Genomic DNA

These are standard human genomic DNA. They can be used in a variety of applications, for instance, genotyping and tissue culture stain identification.

Application: DNA typing, DNA analysis, human identity testing and tissue culture strain identification

Recommended Storage Conditions: -20°C

Name	Cat #	Size
9947A Genomic DNA	DNA HGD-9947A-100	250ng, 10ng/µl
9948 Genomic DNA	DNA HGD-9948-100	250ng, 10ng/µl
K562 Genomic DNA	DNA HGD-K562-100	250ng, 10ng/µl

CE 10X Running Buffer (with EDTA)

MCLAB's CE 10X Running Buffer (with EDTA) has been tested by many labs. It is optimized for use with various ABI's Genetic Analyzers, including 310, 3100, 3130xl, 3730, 3730xl.

Application:

- The CE 10X Running Buffer is used with ABI's POP-4, ABI's POP-6, ABI's POP-7 and MCLAB's NanoPOP™ 4.
- DNA sequencing and DNA fragment analysis
- Optimized for use during capillary electrophoresis on all Applied Biosystems Genetic Analyzers, including 310, 3100, 3130xl, 3700, and 3730, 3730xl.

Recommended Storage Conditions: Room Temperature

Name	Cat #	Size
CE 10X Running Buffer (with EDTA)	RBUF-100	100 ml
CE 10X Running Buffer (with EDTA)	RBUF-500	500 ml

Hairpin DNA & GC rich Sequencing Premix for BigDye® 3.1

MCLAB's Hairpin DNA Sequencing Premix is designed to sequence difficult templates containing hairpin structures and high GC contents.

The example condition:

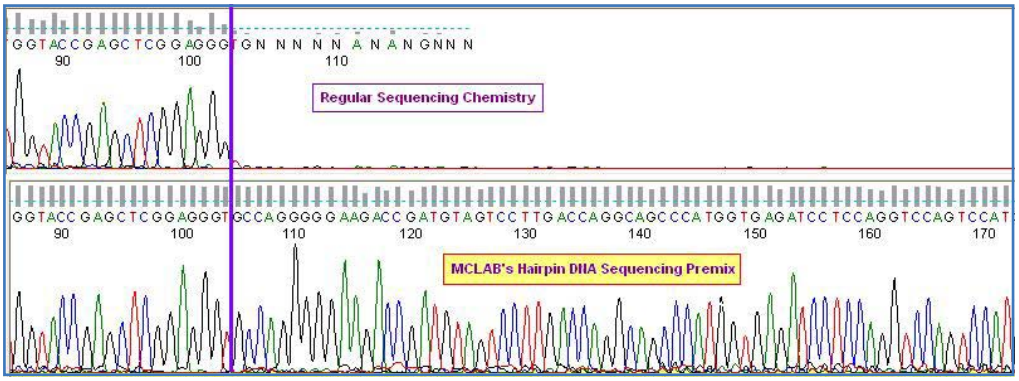
- Hairpin DNA Sequencing Premix 4µl
- DNA template 100ng
- Primer 3.2pmol
- Add ABI's BigDye® 0.125µl
- Add water to final 10µl

Cycle Condition:

- 1 cycle: 98°C for 3 minutes
- 25 cycles: 98°C for 10 seconds; 50°C for 5 seconds, and 60°C for 2 minutes

Recommended Storage Conditions: -20°C

Comparisons: Regular sequencing chemistry vs. MCLAB's Hairpin DNA Sequencing Premix for ABI's BigDye® 3.1



Name	Cat #	Size
Hairpin Premix	BDP-100	1ml, 4µl/rnx

Super-DI ™ Formamide

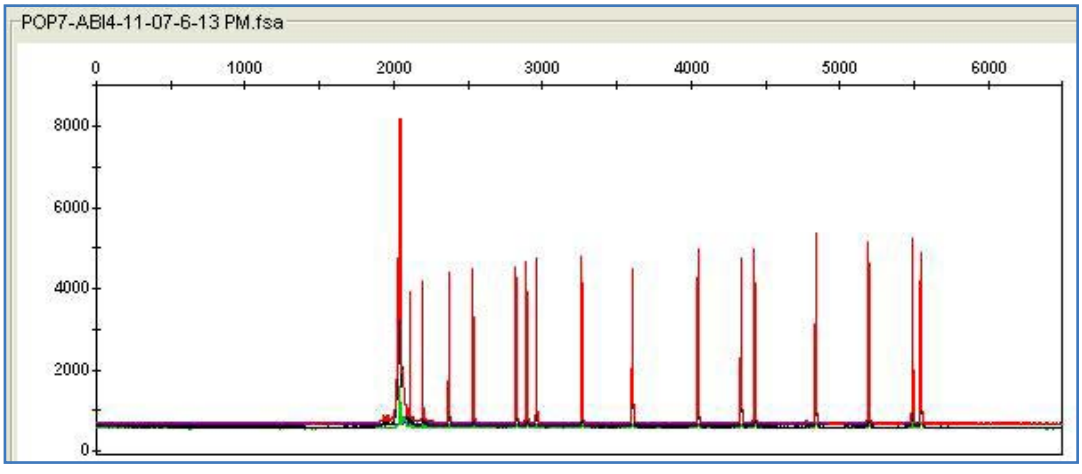
MCLAB's Super-DI™ Formamide is a newly developed loading solution used for DNA denaturation and electrokinetic injection on capillary electrophoresis systems. It is recommended to be used as sample loading solution for all ABI sequencers (3730, 3130, 3100, 310) to ensure sample preservation and resistance to evaporation.

Application: Sample loading solution for all ABI sequencers to ensure sample preservation and resistance to evaporation

Features: Very Stable: remains high signal and excellent performance after storing at 4°C for 6 months.

Recommended Storage Conditions: 4°C.

Example: MCLAB's Super-DI™ Formamide as loading solution



Name	Cat #	Size
Super-Di™ Formamide	SDI-100	25ml

310 Capillaries

310 Capillaries are bare fused silica capillary tubings, 47cm x 50µm (36 cm well-to-read) and 61cm x 50µm (50 cm well-to-read). They are designed and optimized for fragment analysis or sequencing applications on ABI 310 Genetic Analyzer. The capillaries have lower fluorescent background and are very reliable (>100 runs/capillary).

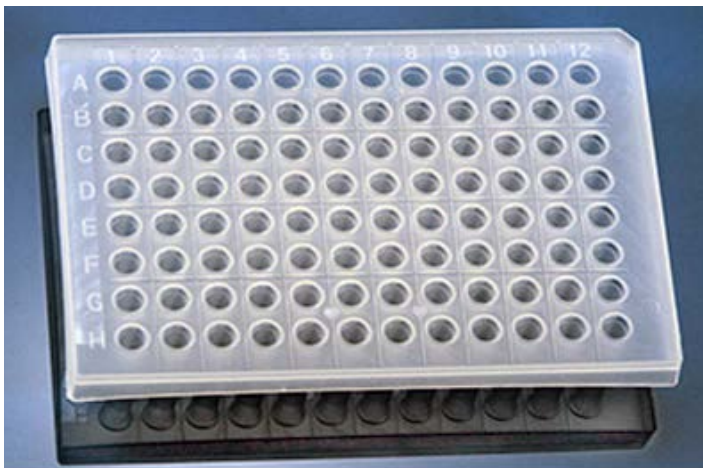
Name	Cat #	Size
310 Genetic Analysis Capillary, 47 cm	CAP-47	5
310 Genetic Analysis Capillary, 61 cm	CAP-61	2

96-well PCR plate with 8-strip Caps

The 96-well plate accompanied with flat 8-cap strip can be used to ship 96-well plate samples for DNA sequencing and other purposes.

The 96 well plates are a cost effective alternative for use in any Applied Biosystems regular or Real-time PCR thermal cycler. They have a very rigid, extra-stabilized frame, and an elevated skirt. Plates are suited for both automatic loading as well as robotic handling. To improve real-Time PCR signal yields, all tubes in this 96-well plate are designed frosted.

The 8-cap strip is the choice of closure for Real-Time PCR, but can also be used in regular cycling experiments. It features an extremely clear flat glass-grade area that equals the performance properties of optical seals. Strips have frosted writing areas at terminal sides.



96-well PCR plate

8-strip caps

Name	Cat #	Size
96-well PCR plate with 8-strip Caps	96P8C-010	10 sets, each has 10 96-well plates (96 x 0.2ml) and 120 8-cap strips for closure of the plates.

MCLAB Products are distributed in Australia by

GeneTargetSolutions

For Research Use Only. Not for use in diagnostic procedures.

Unit 2a / 827 Old Northern Rd DURAL NSW 2158
Ph: (02) 8090 2675
info@genetargetsolutions.com
www.genetargetsolutions.com.au