Pippin Prep 2011: New Applications

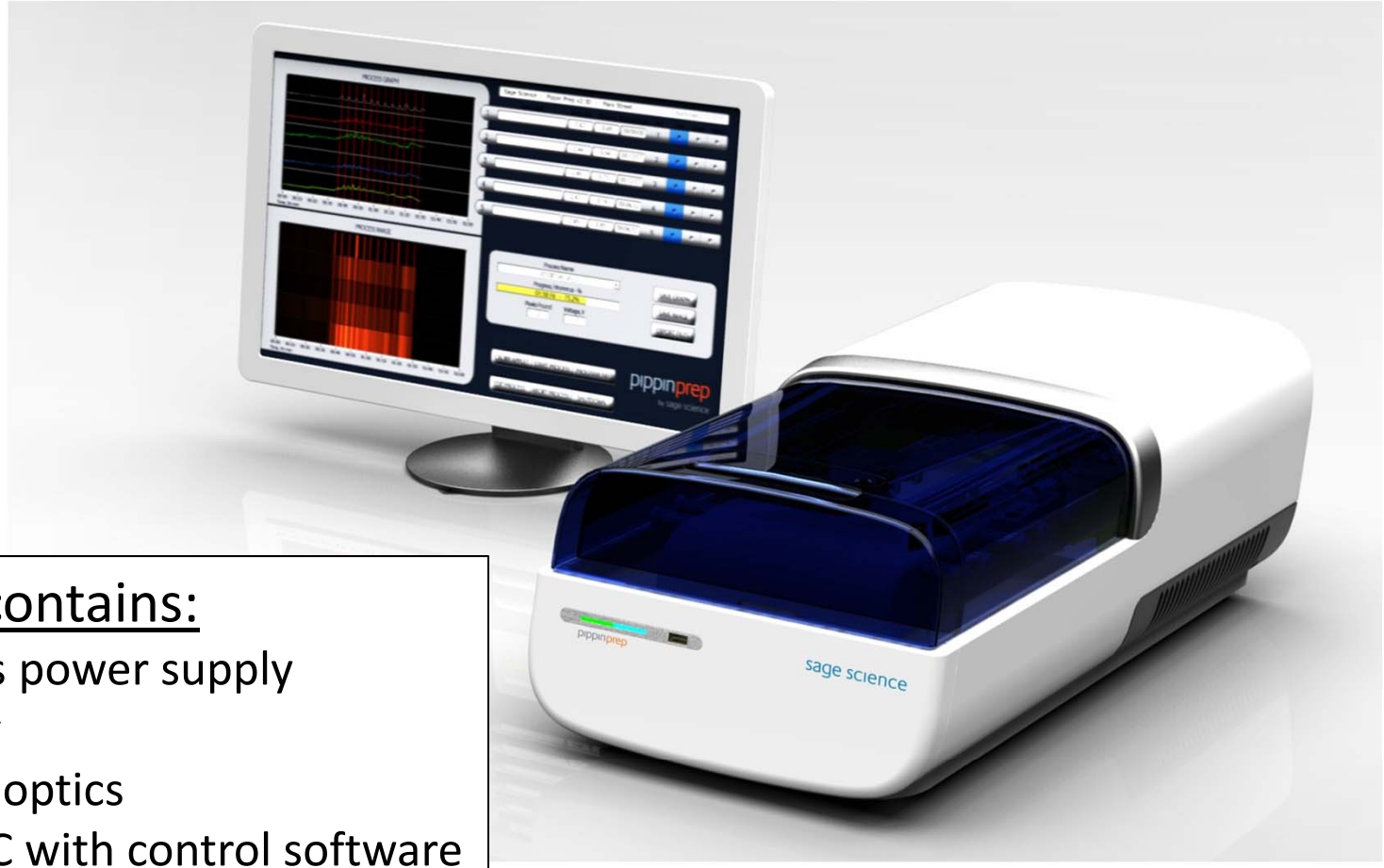
Expanded Cassette Offerings and System Improvements

Chris Boles, Ph.D.
VP & CSO, Sage Science
AGBT 2011

sage science

The Pippin Prep System

Automated Preparative Gel Electrophoresis for NGS

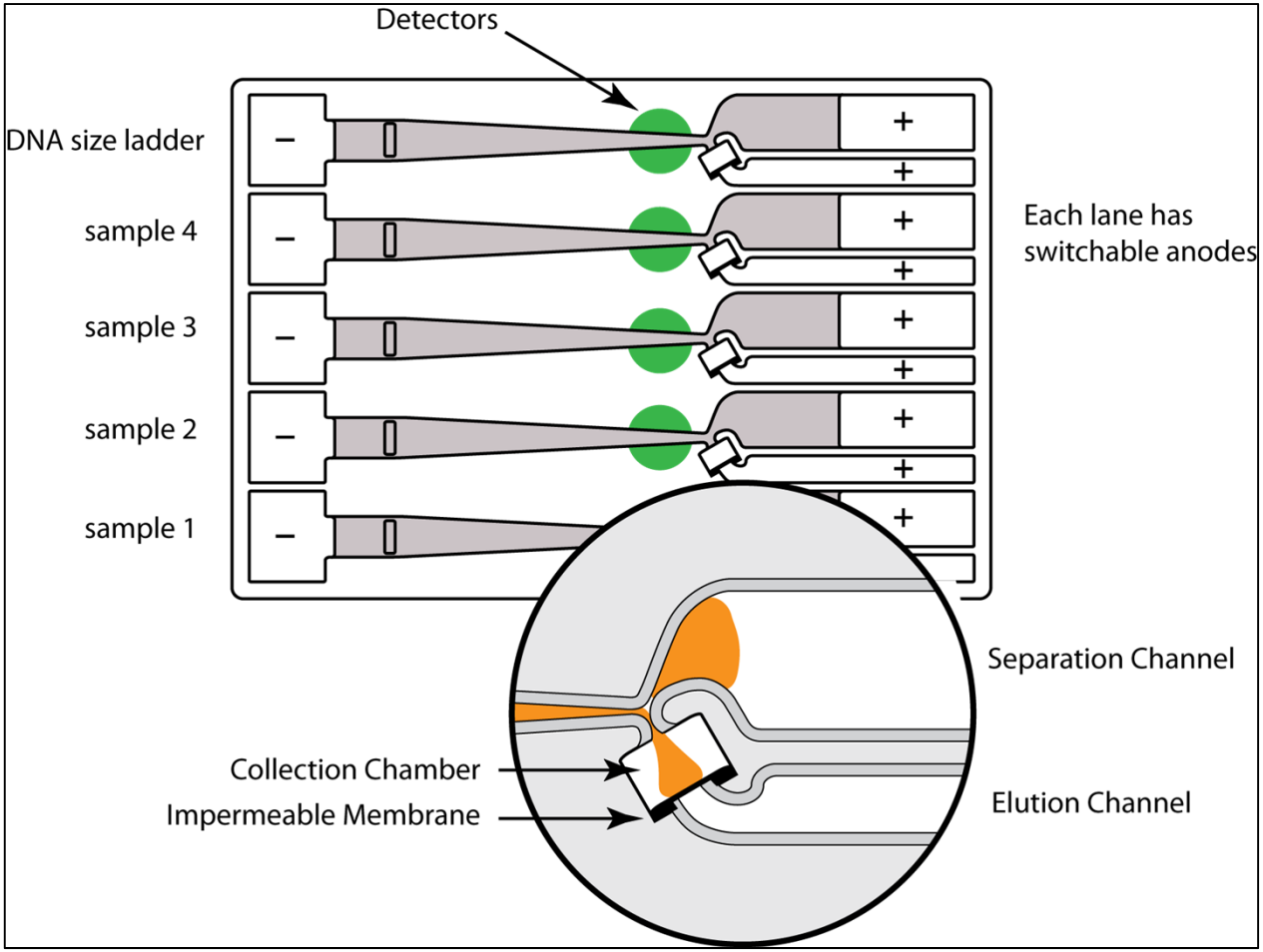


Instrument contains:

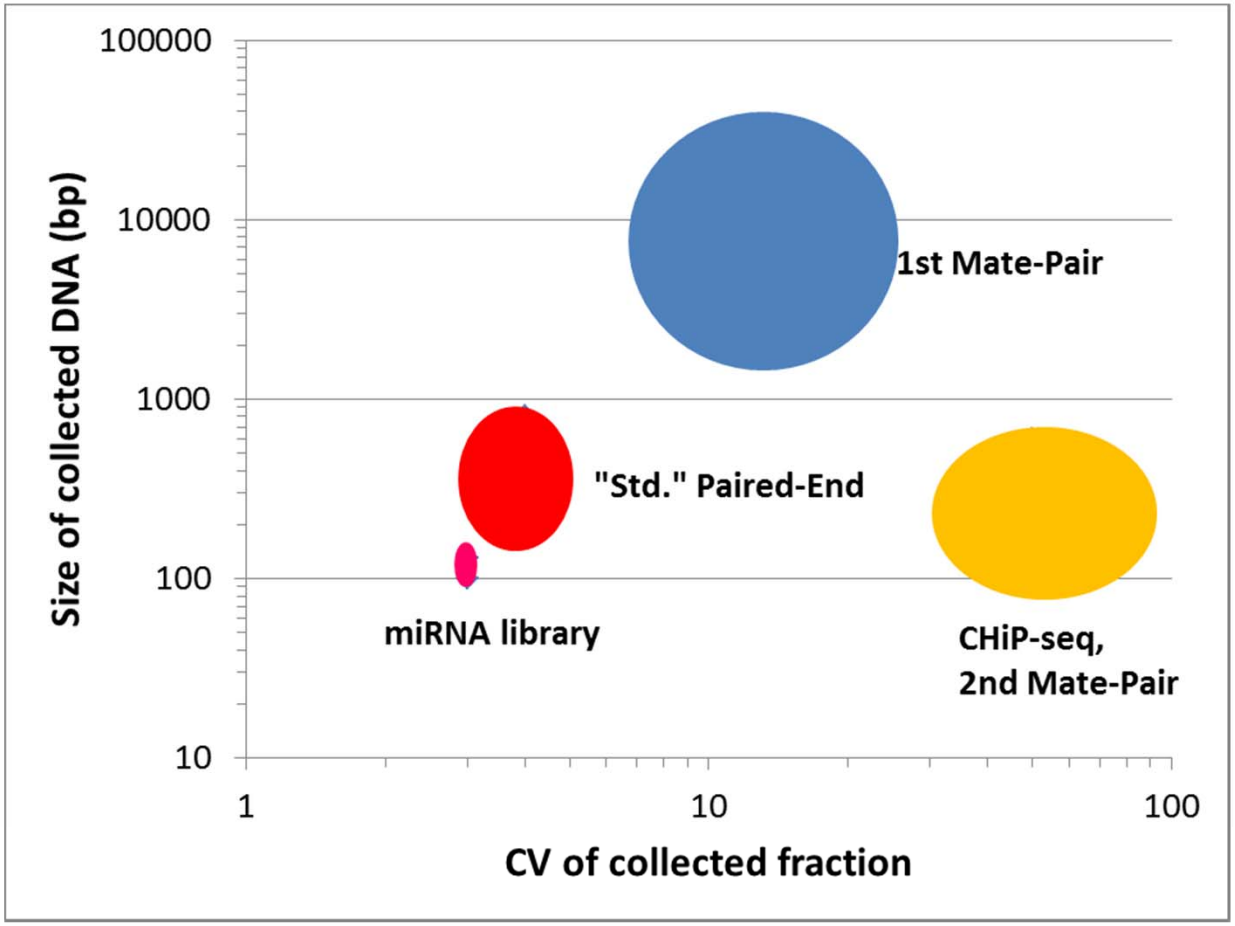
Electrophoresis power supply
Electrode array
DNA detection optics
Single-board PC with control software

The Pippin Prep System

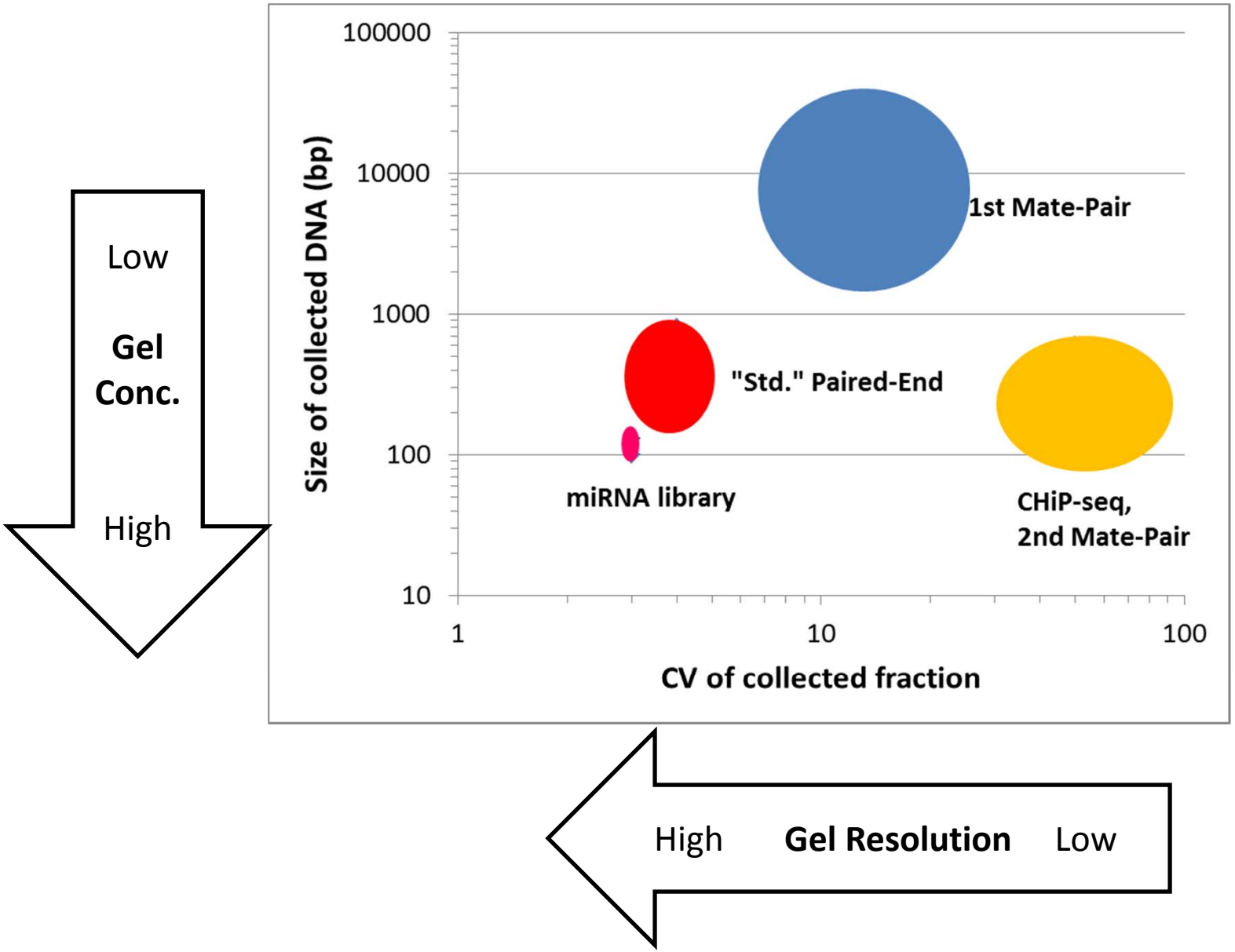
Automated Preparative Gel Electrophoresis for NGS



Size fractionation in NGS Applications



Size fractionation in NGS Applications



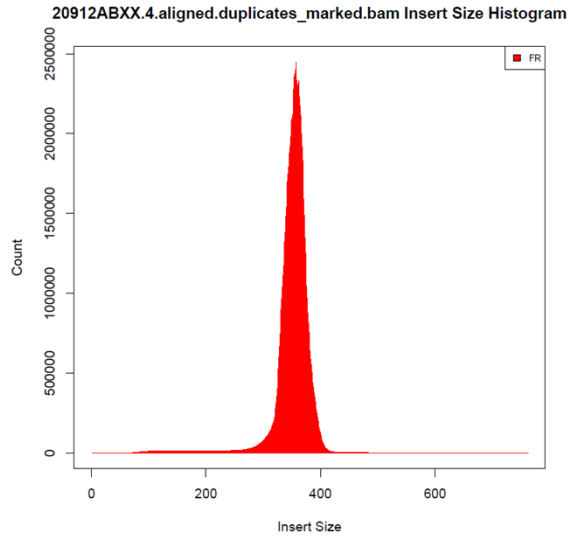
Pippin Prep Products for NGS Applications

Application	Cassette types
PE libraries	2% 1.5% (new, Nov. 2010) 2% Ethidium Free (new Feb. 2011)
Small RNA libraries	3% (new, Feb. 2011)
1st Mate-pair	0.75% (new, Feb. 2011)
CHiP-seq, 2nd Mate-pair	2%, 1.5% cassettes with Closed elution modules (new, Mar. 2011)

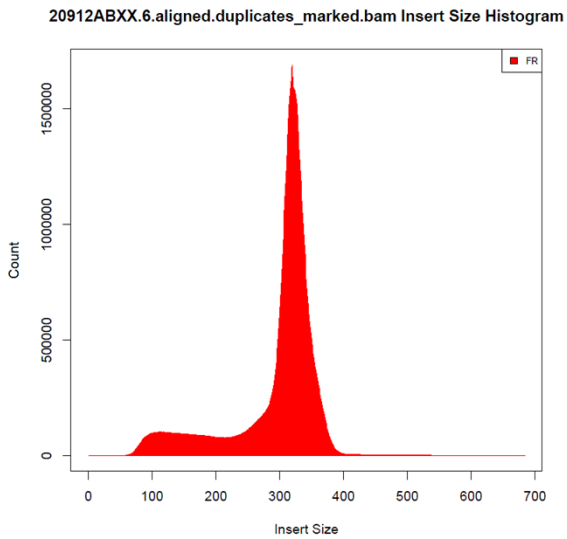
Pippin Prep for PE Libraries: 2% Cassettes

Improvement in PE Library Quality:

Pippin Prep



Manual Gel



Tight insert size.

No LMW “shelf”.

Consistent, high yield.

Work flow and cost efficiency:

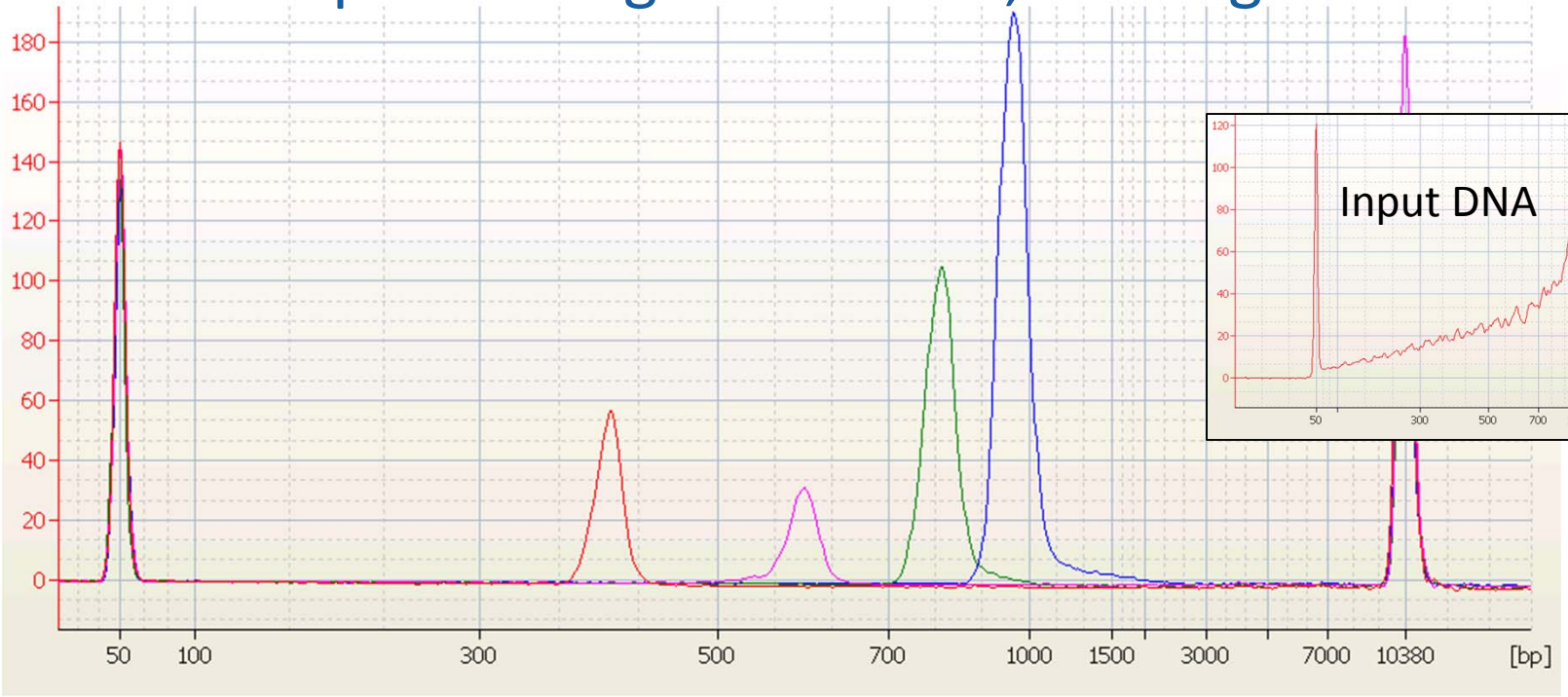
Library production rate doubled per FTE.

10-fold decrease in PCR enrichment cost due to improved yield.

Data courtesy of Broad Institute Sequencing Technology Development Group

Pippin Prep for PE Libraries: 1.5% cassette

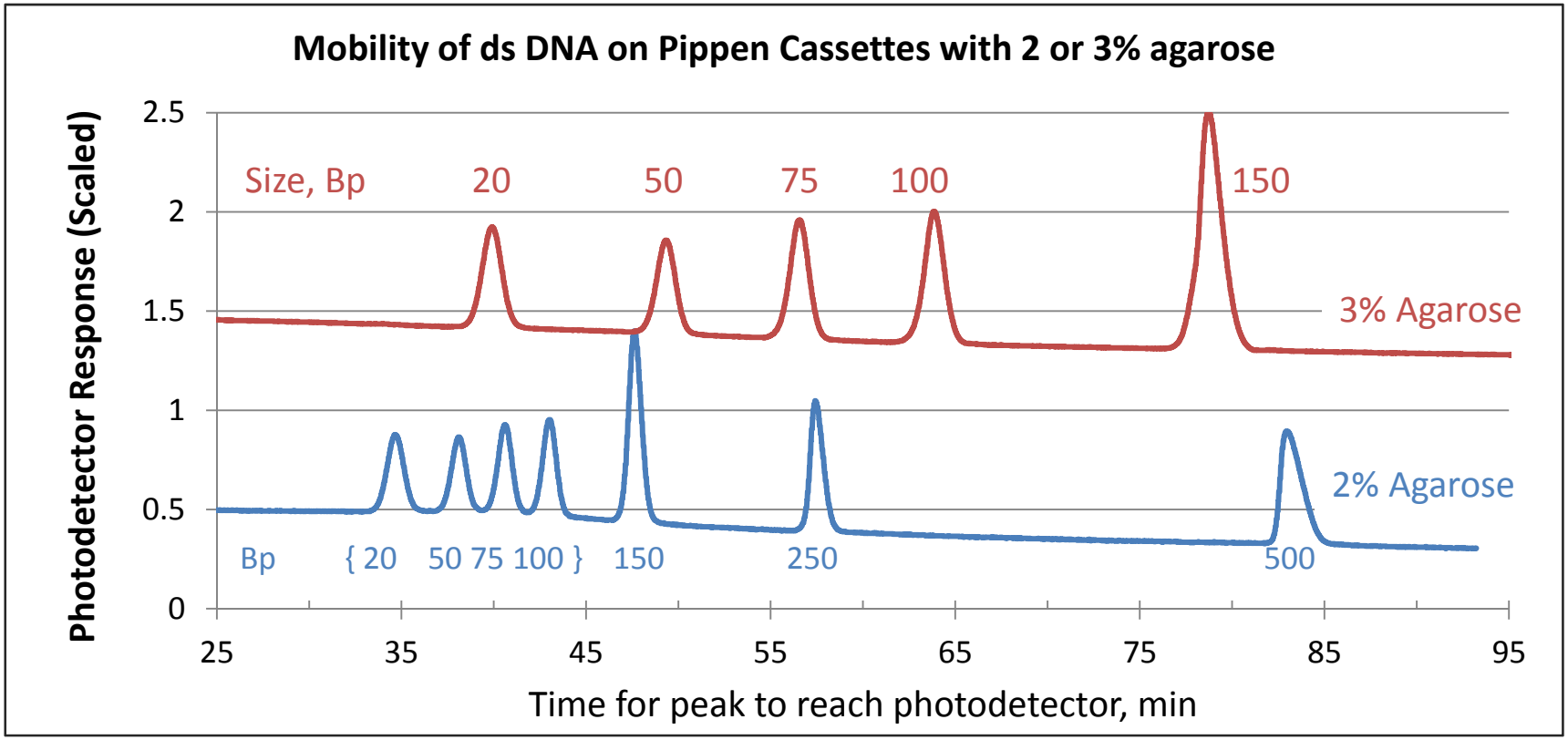
Expands range to ~1.5 kb, with tight CVs.



Programmed	Actual	CV (%)
400 tight	408(364-454)	**3**
600 tight	598 (545-674)	**3**
800 tight	811(690-973)	**5**
1000 tight	977(851-1361)	**6**

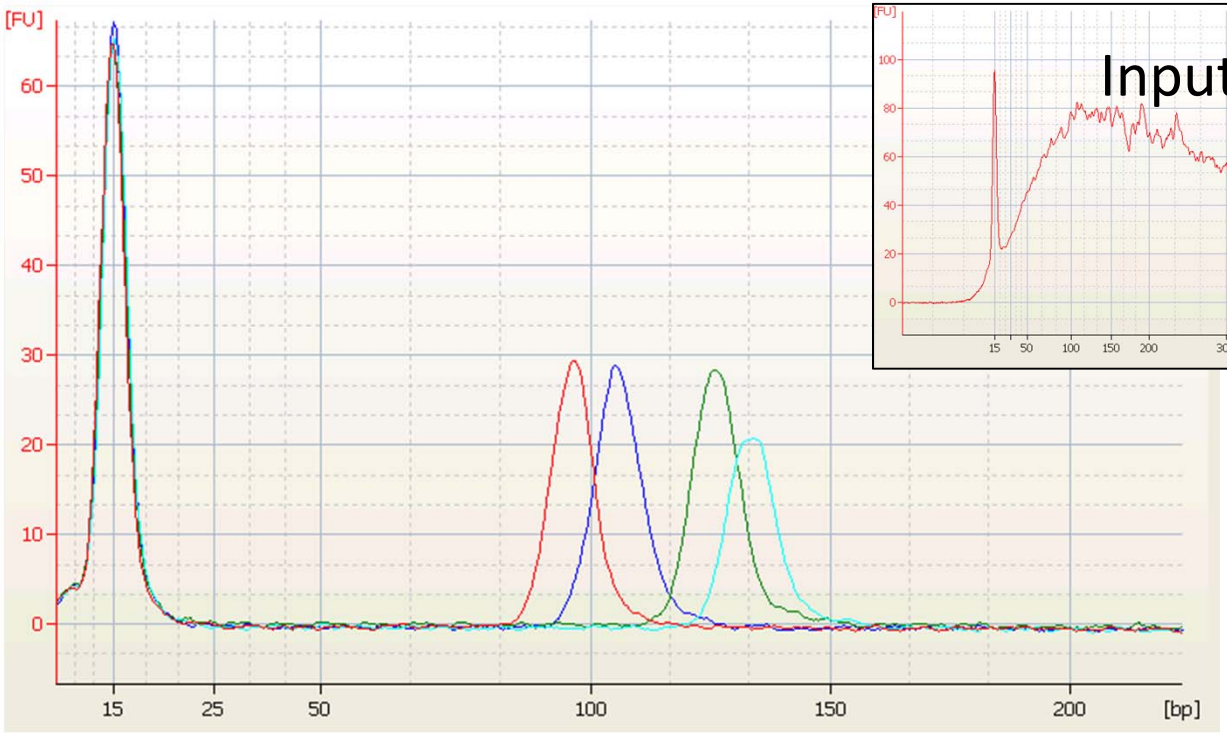
Pippin Prep of miRNA libraries: 3% cassette

Optimal resolution in 50 – 200 bp range.



Pippin Prep of miRNA libraries: 3% cassette

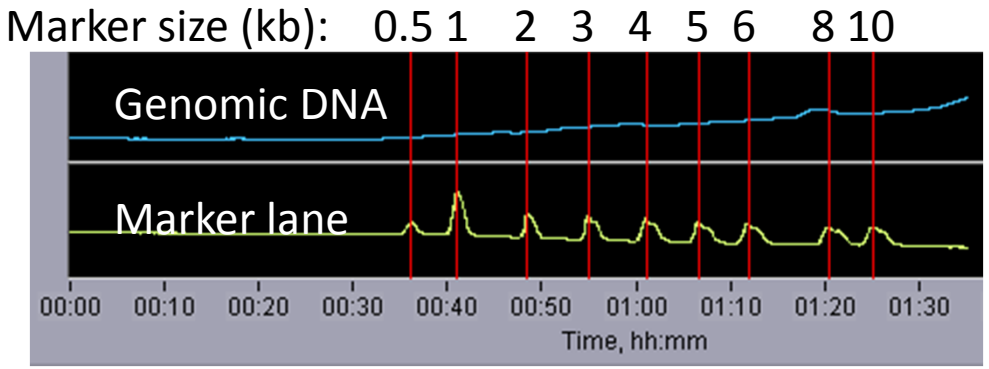
Performance on gDNA samples



Programmed	Actual	CV(%)
90 tight	97 **(82-114)**	5
100 tight	106**(90-129)**	5
120 tight	127 **(109-144)**	4
130 tight	134 **(115-149)**	4

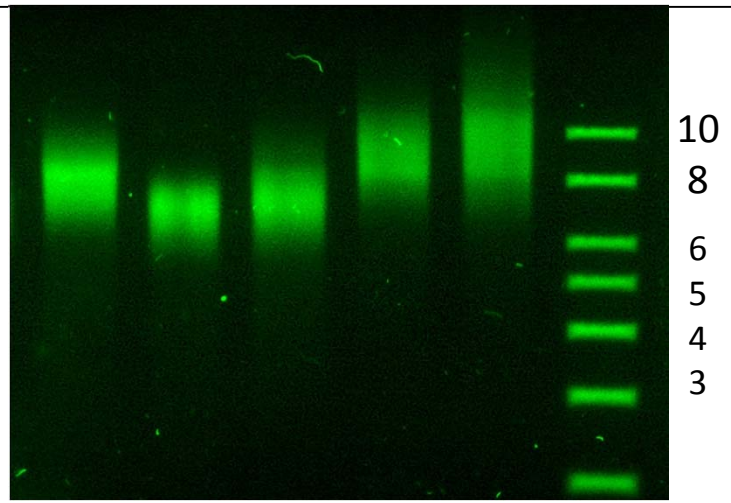
Pippin Prep 0.75% Cassettes for Mate-Pair Libraries

Size range: 1 - 10 kb
Run times: 0.6- 1.5 hr.



Size (kb):	8.2	7.1	7.4	9.1	9.6
CV:	12%	10%	12%	13%	16%

5 µg/lane input:
CVs ~10-16%



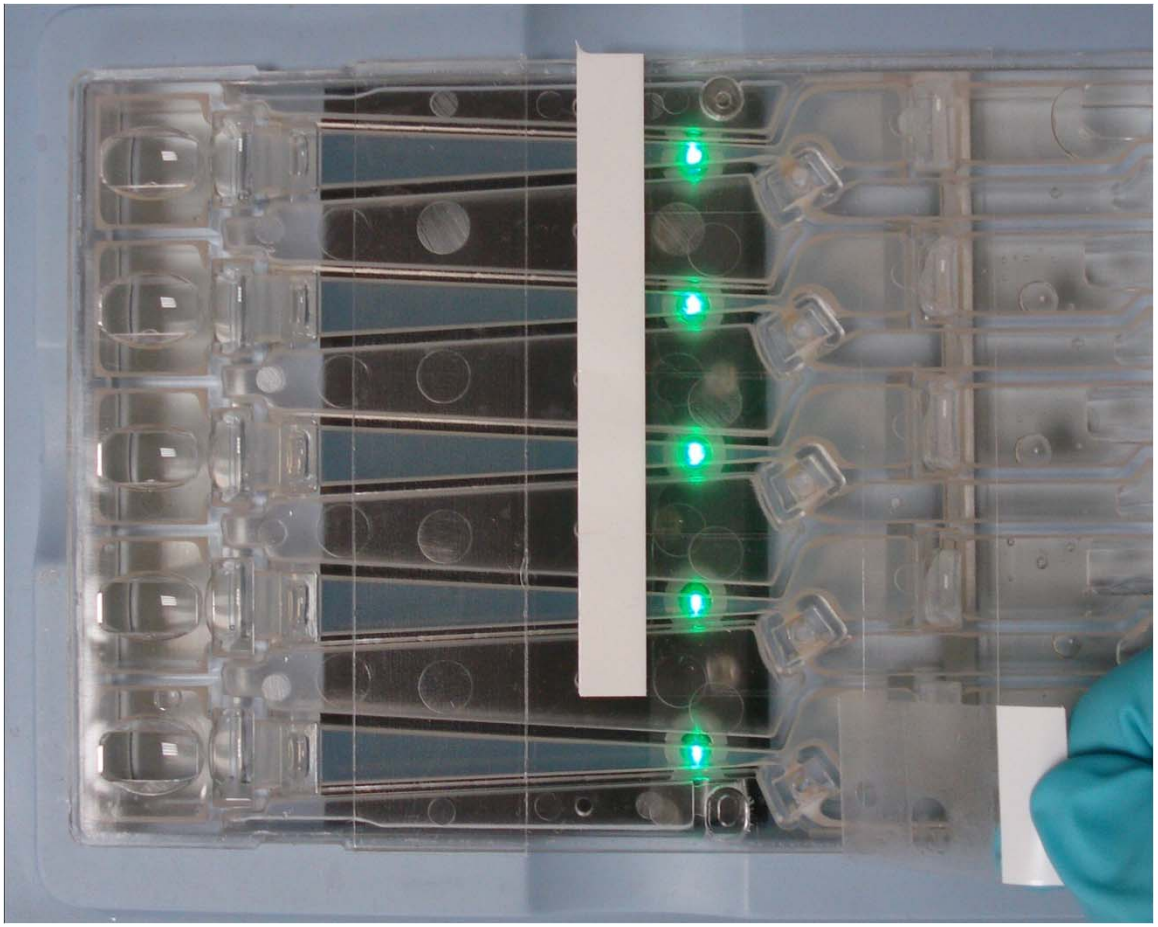
Pippin Prep for CHiP-seq

(and other broad size range applications)

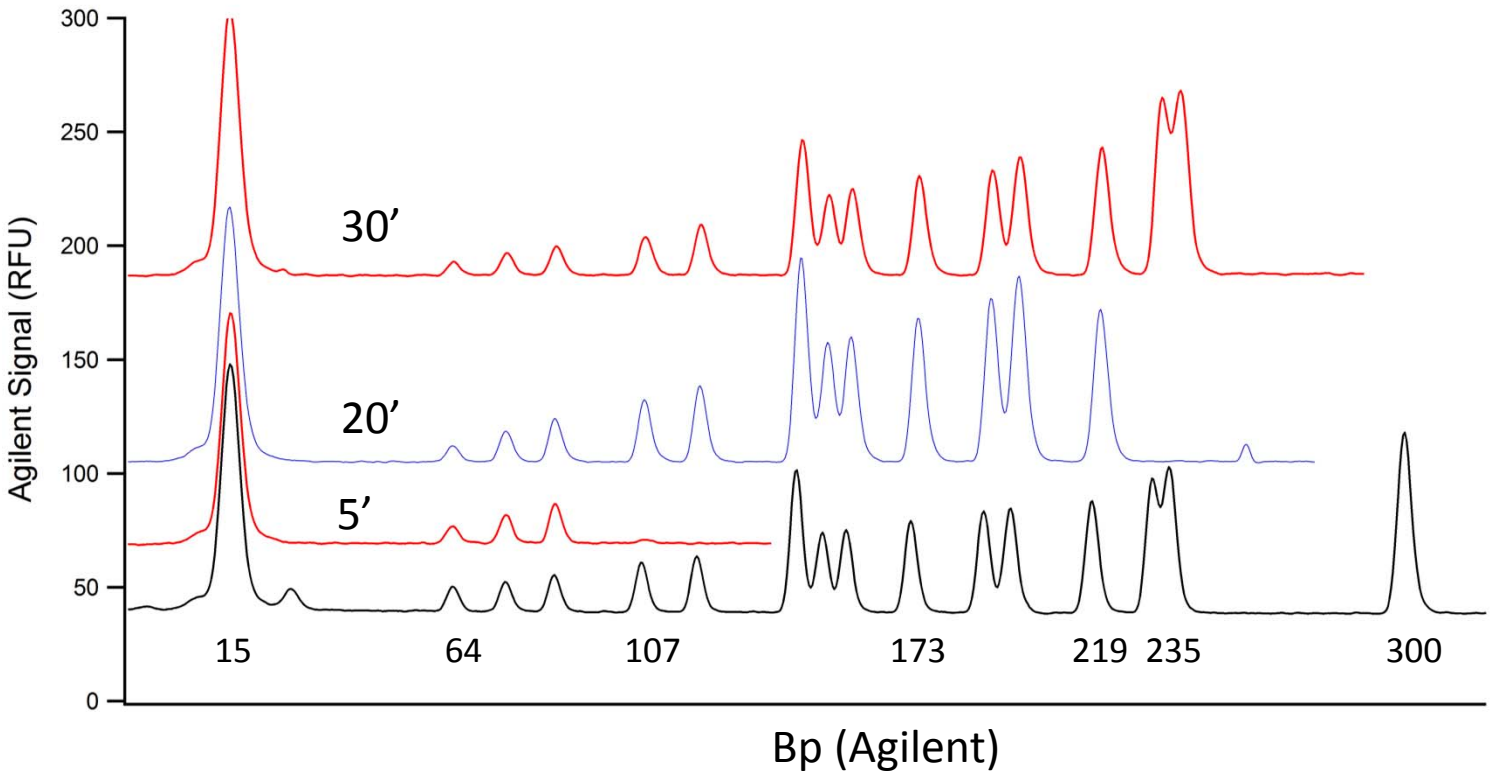
<u>Closed elution modules</u>:
Prevent electroosmotic flow into elution module.

Elution volume remains fixed at 50 µl.

Permits long elution times without overflow or manual intervention.

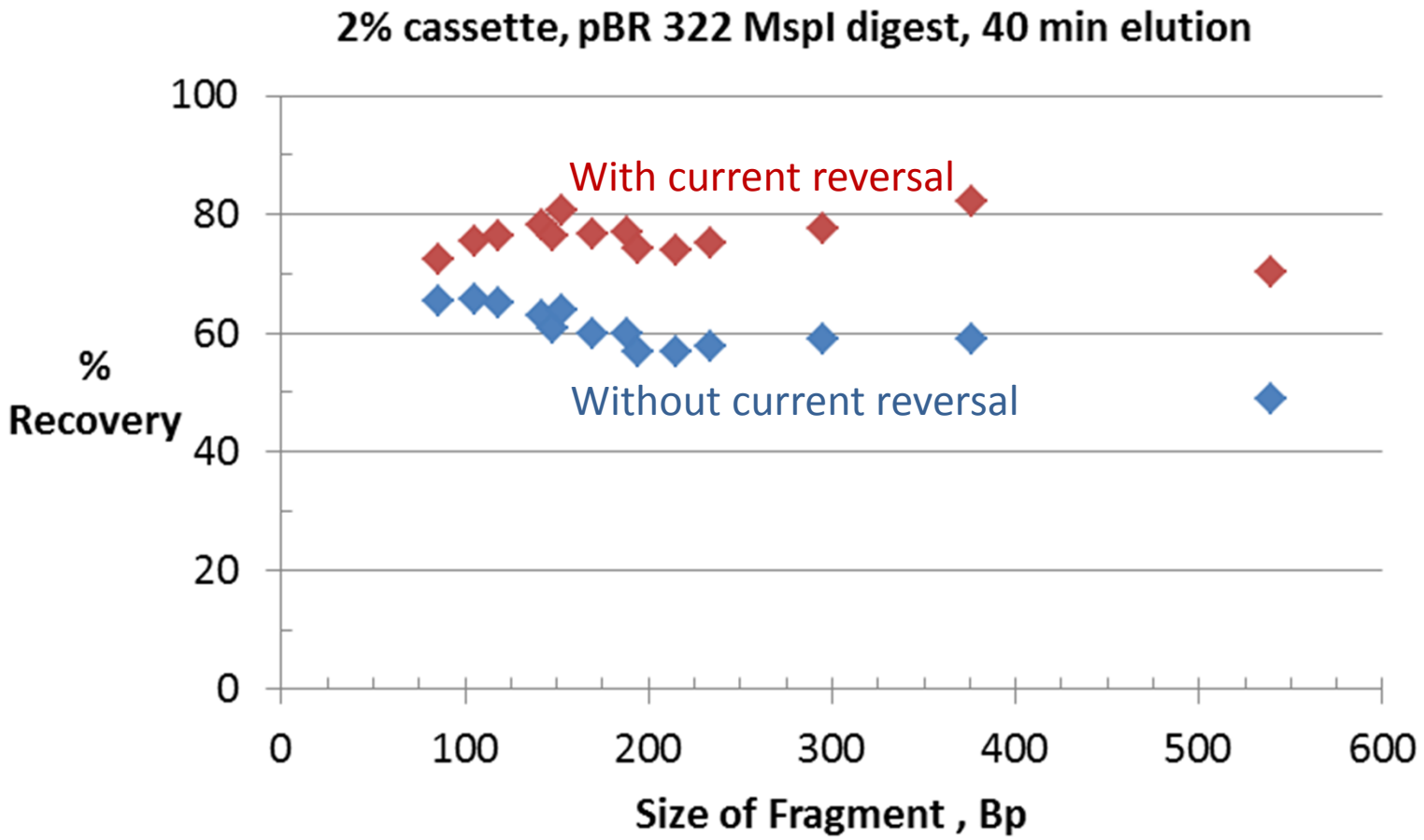


Pippin Prep for CHiP-seq: Long Elutions



Agilent 7500 data of DNA eluted from Pippin Cassette
pBR322 MspI digest run on 2% cassette, with elution times
From 5 – 30 minutes.
DNA analyzed on Agilent 7500, with digest as std

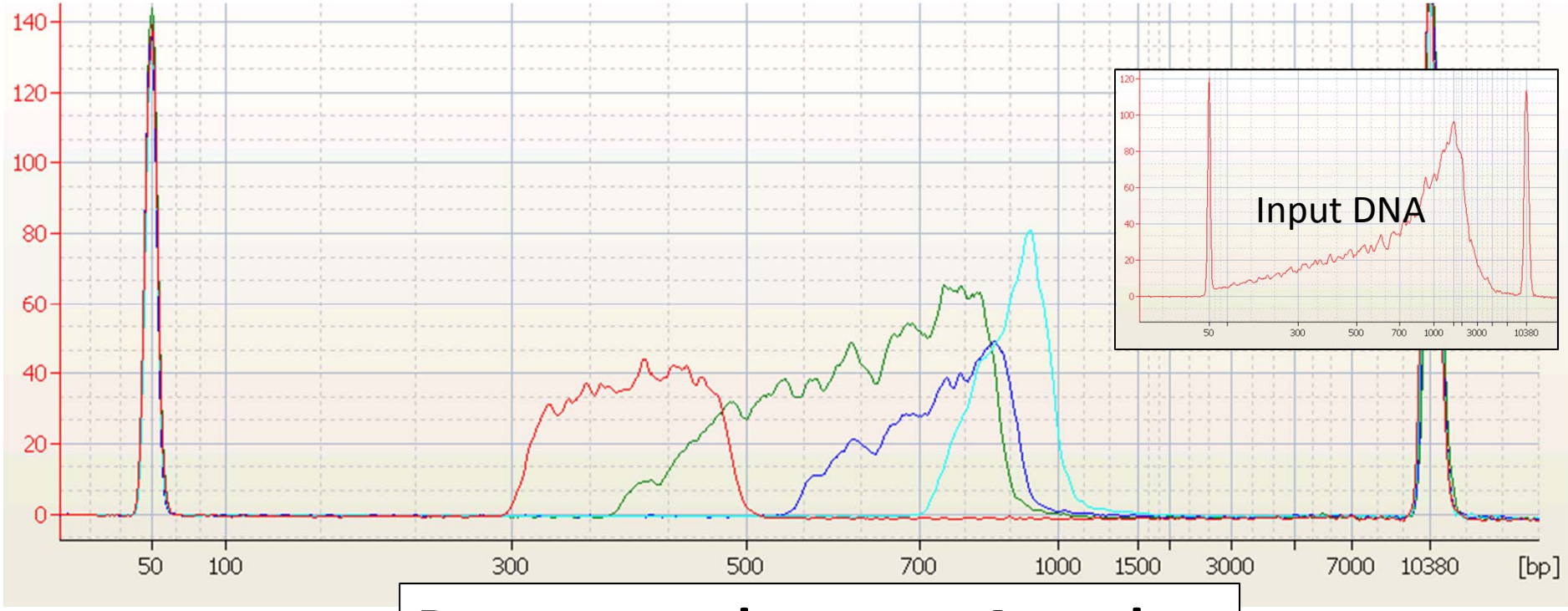
Pippin Prep for CHiP-seq: Consistent high recoveries



Short current reversal in elution module before sample removal boosts recovery to >60%

Pippin Prep for CHiP-seq

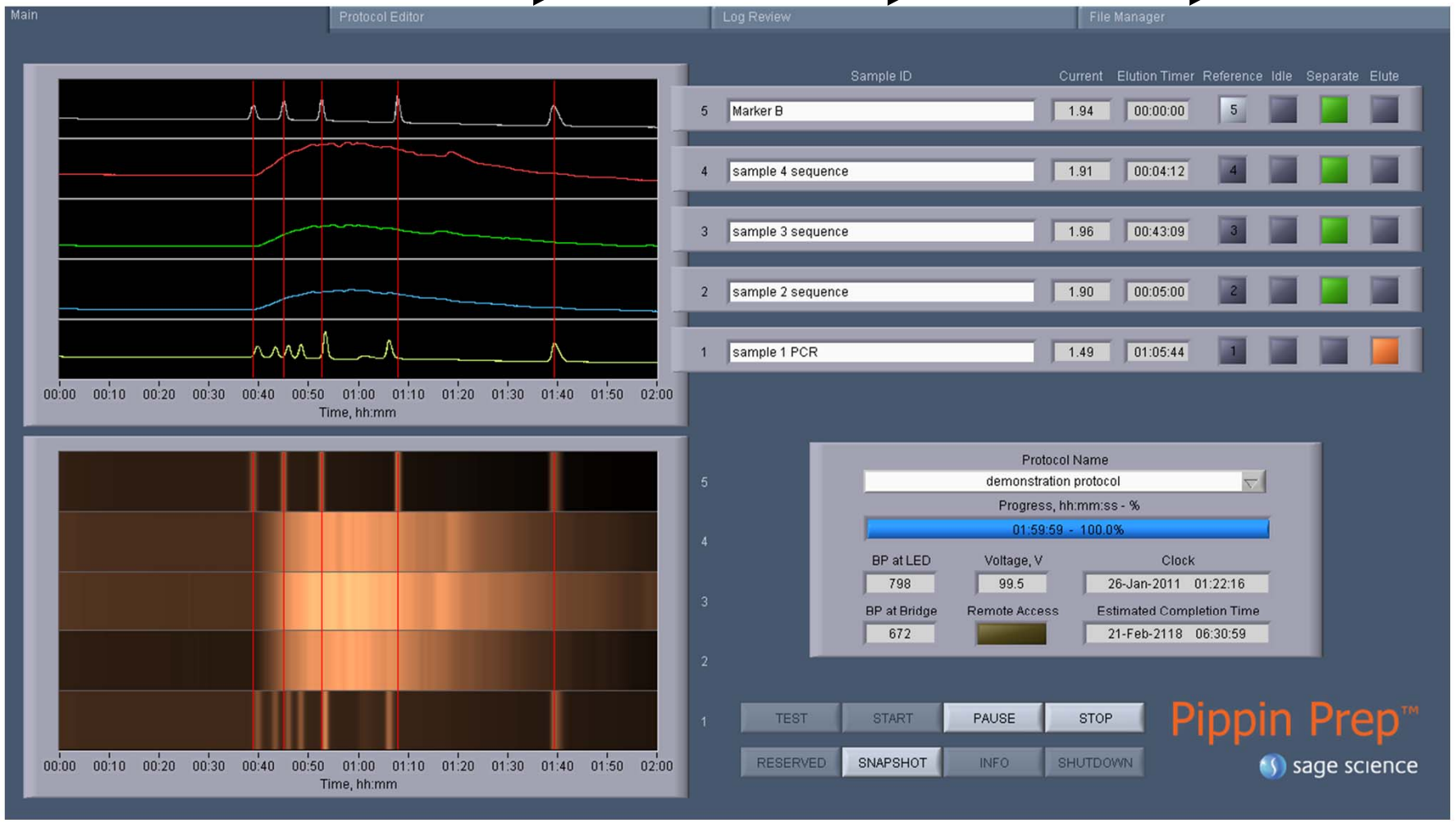
Broad collections in 1.5% cassette (sealed elution modules)



Programmed	Actual
300-500	291-523
400-900	376-925
550-950	535-976
700-1100	683-1182

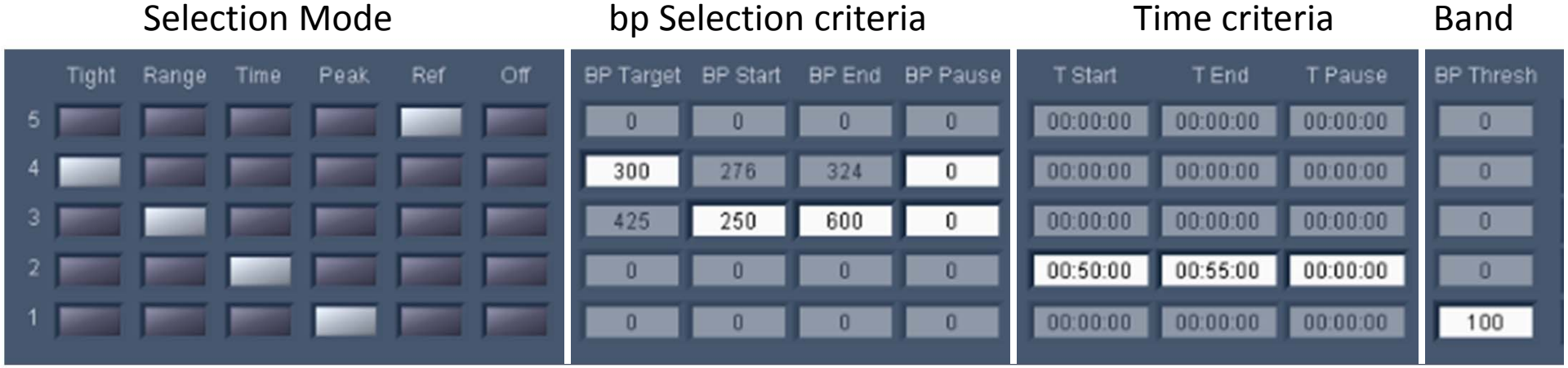
New Pippin Prep System Software: Tabbed Browser Format

With run in progress, user can **edit** other protocols, **review** old runs, **manage** log files.



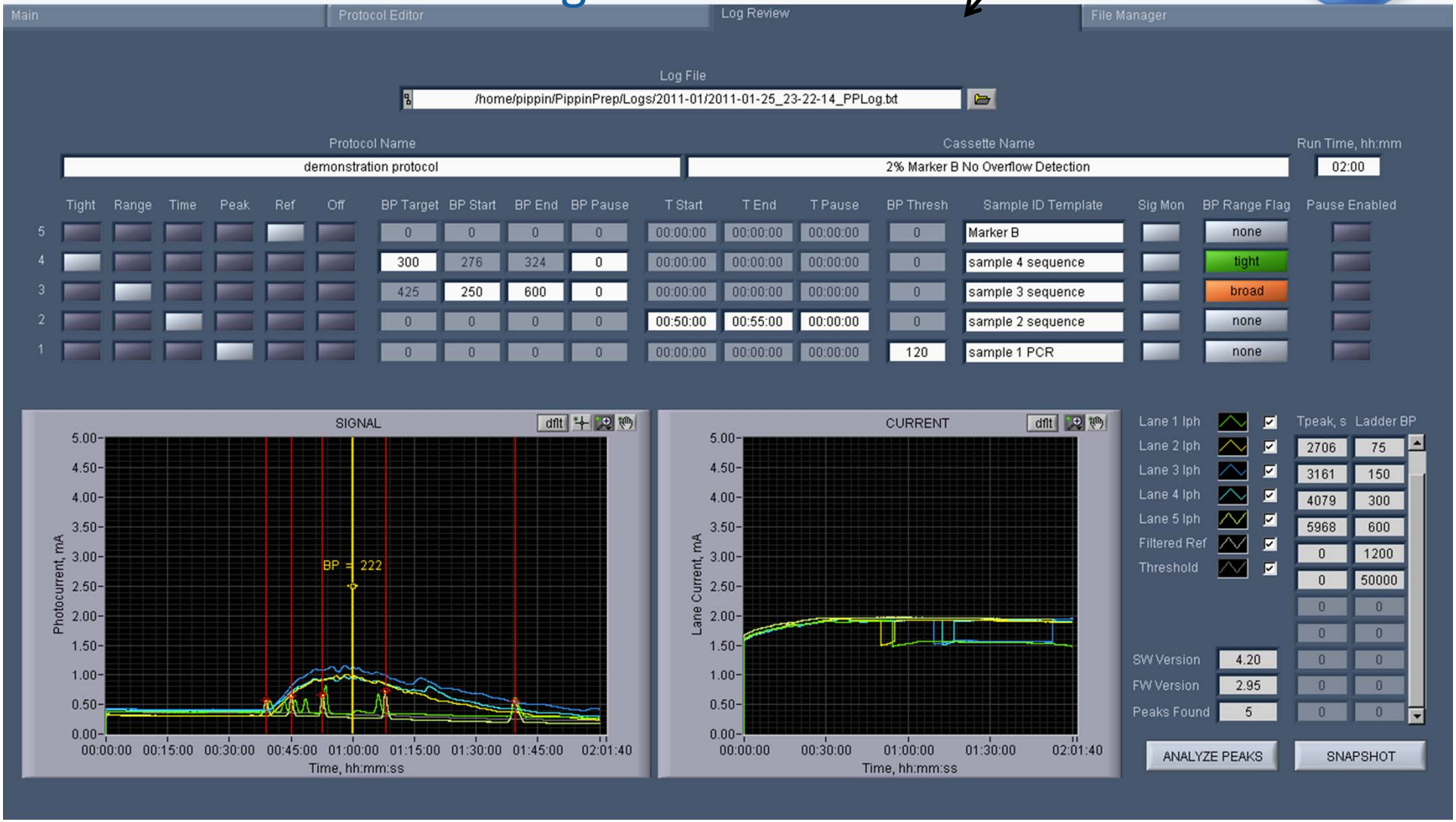
Home screen shows **time to completion**, **bp position at LED** and **branch point**.

New Pippin Prep System Software:
New Collection Modes and Protocol Editor



Tight = Capture tightest band centered on BPtarget value
Range = Collect between BPstart and BPend value
Time = Collect between Tstart and Tend
Peak = Collect first band encountered in sample after BP threshold value

New Pippin Prep System Software: Log File Review Tab



Shows **<u>protocol details</u>** (top), interactive plots of **<u>optical</u>** and electrophoresis **<u>current</u>** data, while new run is in progress.

Pippin Prep 2011: Summary

User-validated performance in PE library construction

Benefits in throughput, process cost, and library quality

New offerings for Q'1 2011 expand system size range: **75 - 10,000 bp**

A cassette (or two) for almost every NGS application:

Libraries for **PE reads, miRNA, mate-pair, CHiP-seq**

Please visit us at poster **#27** for more details and questions. Come and see the Pippin Prep in our **Lanai suite: #292**.

Sales inquiries: paul.ventura@sagescience.com
Technical inquiries: chris.boles@sagescience.com, sadaf.hoda@sagescience.com
Company inquiries: gary.magnant@sagescience.com
Other info and contact information on our website: www.sagescience.com