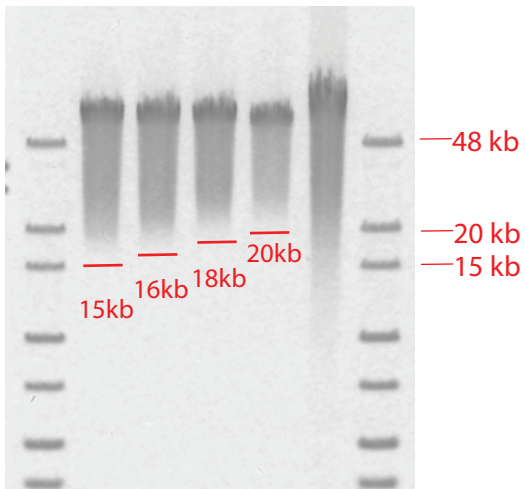
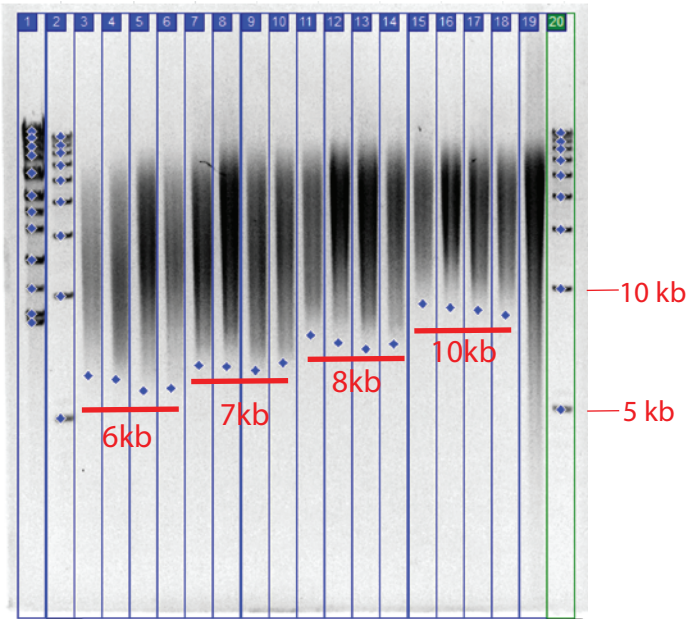


Sheared DNA Samples

The gel image below show sheared genomic DNA samples run on the BluePippin using high-pass protocols.



** These data are not intended to imply guaranteed results or performance. This product is intended to demonstrate that the BluePippin is functioning as expected, and that proper operational technique is being used. Users should refer to the Operations Manual for performance specifications.*

BluePippin™

Control DNA

For Testing and Validation of
0.75% Agarose Gel Cassettes

For High-Pass protocols

Gel Cassette Kit Numbers:

BLF7510

PAC20KB

High-Pass Protocols:

>6-10 kb Version3

>15-20 kb

What is Enclosed

Control DNA for High-Pass protocols consists of a DNA ladder with 16 size markers between 0.5 - 36 kb (16 sample loads [1.25µg/40µl] in 680 µl total volume). With High-Pass protocols, users set a threshold (between 6-10 kb or 15-20 kb) in the BluePippin software. DNA above that threshold will be collected, and lower molecular weight DNA will be filtered out from the genomic sample.

To Use

1. Use the PAC20KB (Marker S1) or BLF7510 (Marker S1) agarose gel cassette .
2. Carefully follow the cassette preparation and sample load instructions that are outlined in the BluePippin Operations manual, BLF7510 Quick Guide, or the "High-Pass Guide for SMRTbell templates for the PacBio RS system" .
3. Load the "0.75% DF Marker S1 high-pass 6-10kb vs3" or "0.75% DF Marker S1 high-pass 15-20kb" cassette definition in the BluePippin software protocol editor and enter one or more of the size selection parameters shown below.
4. Pippette 40µl of control DNA into a sample well or wells and load marker S1 into the well for the designated reference lane.
5. Analyze the collected fractions on pulsed-field slab gel (using Pippin Pulse) for sizing, and/or Qubit® Fluorometer and Quant-iT™ HS dsDNA reagent for quantitation to assess yield.

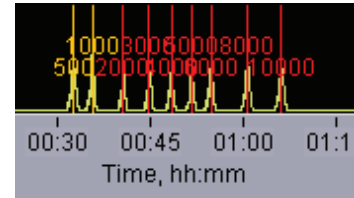
Sample protocol for >6-10 kb high pass (marker is in lane 1)

	Tight	Range	Time	Peak	Ref Lane	BP Target	BP Start	BP End	BP Pause
5					1	28000	6000	50000	0
4					1	28500	7000	50000	0
3					1	29000	8000	50000	0
2					1	30000	10000	50000	0
1					1	0	0	0	0

Sample protocol for >15-20 kb high pass (marker is in lane 1)

	Tight	Range	Time	Peak	Ref Lane	Target *	Start *	End *	Pause *
5					1	35000	20000	50000	0
4					1	34000	18000	50000	0
3					1	33000	16000	50000	0
2					1	32500	15000	50000	0
1					1	0	0	0	0

Typical Results



Marker S1

At the end of a run, marker peaks will be detected in designated marker lane in the BluePippin main screen software display.

NEB

1 kb

Extend

Ladder

15 kb

16 kb

18 kb

20 kb

High-Pass threshold setting

6 kb

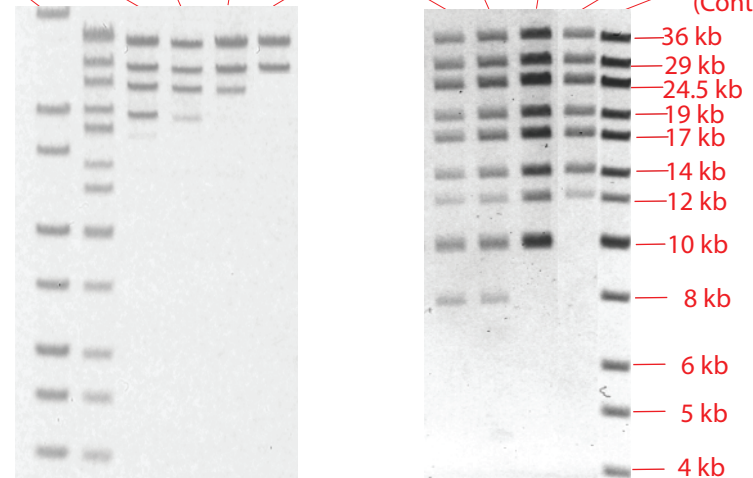
7 kb

8 kb

10 kb

None

(Control DNA)



The gel images above shows expected size selections of control DNA CHP7504, at 6-10kb and 15-20kb thresholds, when compared to the non-selected marker. 10µl of the total 40µl elution was loaded on the gel.

The analytical gel was run with a Pippin Pulse using the 10-48kb pre-set protocol and run for 15 hours. 0.75% SeqKem Gold agarose from Lonza was used.