Control DNA - IS
For Validation of 1.5% Gel Cassettes with Internal Standards

collects targets between 250 bp - 1500 bp

Item# CIS1504

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

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Pippin cassettes and instruments are functionally tested using restriction digests of genomic DNA from E. coli. For each cassette type, a different restriction digest is used, chosen so that size distribution of the digested DNA closely matches the useful fractionation range of the cassette, without any significant peaks or discontinuities. Following restriction digestion, the control DNA is purified by phenol:chloroform extraction, dialyzed, and diluted into Pippin electrophoresis buffer.

Enclosed is one tube of gDNA digest with sufficient volume for 16 sample loads. After internal standards are added, the final DNA load amount is 2 µg per sample, the same amount used for calibration and ongoing quality control.

Control DNA is useful to test, refine, and troubleshoot Pippin size fractionation protocols. It can also be used to check system performance.

To Use

1. Pippette 30 µl of gDNA digest and transfer to reaction tube.
2. Add 10 µl of internal standard to the tube (Marker K for Pippin Prep CDF1510, or Marker R2 for BluePippin BDF1510).
3. Mix samples thoroughly (vortex mixer). Briefly centrifuge to collect.
5. Pippette 40 µl of the gDNA/marker mix into a sample well.

QC protocol for 1.5% agarose cassettes

Cassettes are tested using “Tight” mode with the following target values. Extracted samples are run on an Agilent Bioanalyzer using a DNA 7500 chip. The analysis volume is 1 µl from a 40 µl elution volume (1:40 dilution).

<table>
<thead>
<tr>
<th>Tight</th>
<th>Range</th>
<th>Time</th>
<th>Peak</th>
<th>Ref Lane</th>
<th>BP Target</th>
<th>BP Start</th>
<th>BP End</th>
<th>BP Pause</th>
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<td></td>
<td></td>
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<td>1832</td>
<td>0</td>
</tr>
</tbody>
</table>

Typical Results

Since internal standards are used with dye-free cassettes, the sample DNA will not be optically detected. Marker peaks will appear in each lane:

Target = 250 bp
Ave Size = 255 bp
Conc. [ng/ul] = 3.22

Target = 400 bp
Ave Size = 402 bp
Conc. [ng/ul] = 1.78