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APPLICATION NOTE

Bioruptor® DNA shearing for Agilent SureSelect library preparation: a perfect combination

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Cologne Center for Genomics

The Cologne Center for Genomics (CCG), headed by Prof. Dr. Peter Nürnberg, is a central core facility of the University of Cologne which serves both research groups inside and outside the university. To stay abreast of recent developments in DNA sequencing, the center operates several different next-generation sequencing (NGS) devices and employs a highly skilled staff capable of performing a variety of challenging NGS applications.

DNA shearing is an important step in next-generation sequencing applications. Therefore, a fast, reliable, and high-throughput method is essential. The Bioruptor® NGS* (Diagenode) system fulfills these requirements.

As an example, the CCG implemented an automated library preparation system, the Agilent Technologies BRAVO System, to prepare and enrich DNA in a 96-well format. The protocol "SureSelect XT Automated Library Prep and Capture System (SureSelect^{XT} Automated Target Enrichment for Illumina Paired-End Multiplexed Sequencing; Protocol Version F.0)" requires DNA shearing to get small fragments (150-180 bp) in a small volume (50 µl) using the "Sample Preparation 200 ng DNA" protocol. The Bioruptor® NGS (Diagenode) was essential for this sample preparation workflow. We sheared 200 ng DNA in 50 µl using the Bioruptor® NGS with 0.1 ml Bioruptor® Microtubes (Diagenode, Cat. No. C30010015) and the 0.1 ml tube holder and tube adaptor (Diagenode, Cat. No. B01200041). DNA was diluted to 4 ng/µl with Qiagen EB Buffer. After an equilibration time of one hour at room temperature the samples were quickly vortexed and centrifuged. Prior to sonication the samples were stored on ice for 10 minutes. The DNA was sheared using the following Bioruptor® settings: 30 seconds on/30 seconds off, 60 cycles, followed by a short centrifugation after each round of 15 cycles. The size distribution was analyzed on an Agilent Technologies 2200 TapeStation system. An example result is shown here:

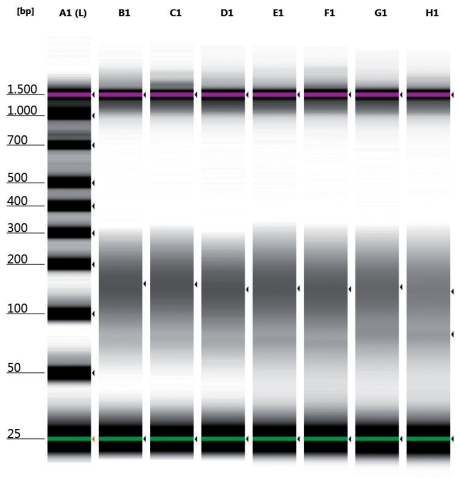

Figure 1: Efficient and reproducible DNA shearing
200 ng of high molecular weight DNA was sheared for 4 x 15 cycles of 30 sec ON/OFF with the Bioruptor® NGS.

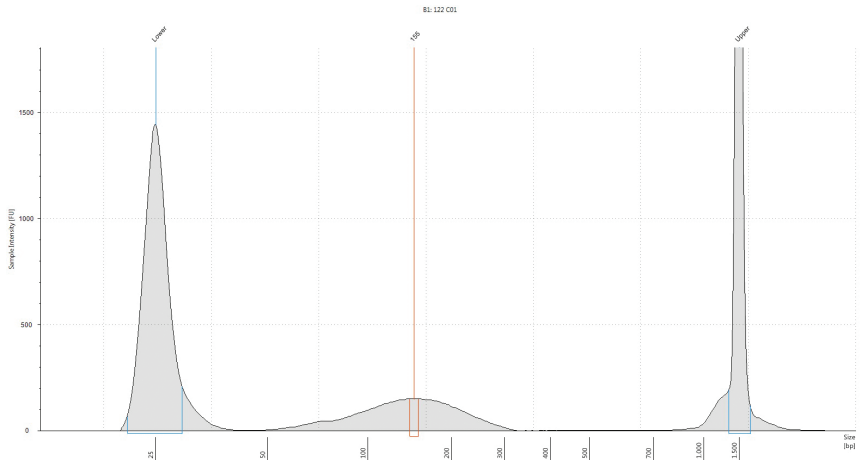

Figure 2: Peak analysis of sample B1

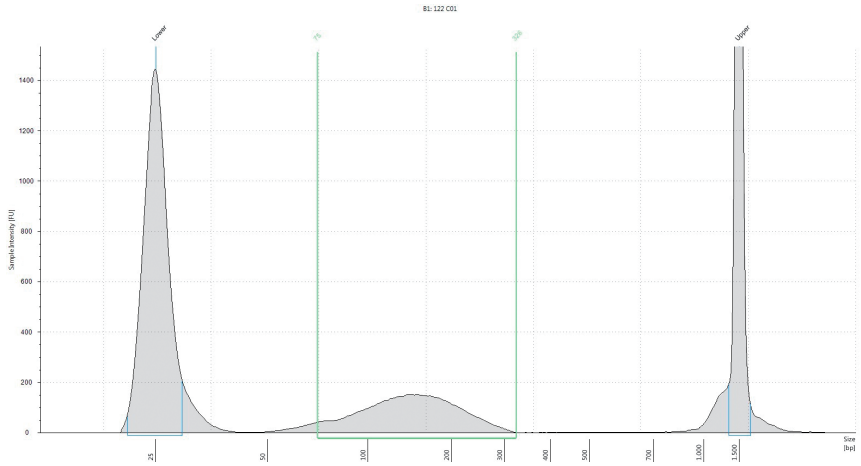

Figure 3: Region analysis of sample B1

Sample	From [bp]	To [bp]	Average size [bp]	Conc. [pg/µl]	Region molarity [pmol/l]	% of Total
B1	75	350	157	2.95	32.5	66.42
C1	75	350	159	3.08	33.6	66.45
D1	75	350	149	2.82	32.5	65.12
E1	75	350	156	3.05	34.3	64.70
F1	75	350	151	2.89	33.4	60.92
G1	75	350	150	2.61	30.4	62.56
H1	75	350	149	2.17	25.6	58.12

Table 1: Region analysis of sample B1 - H1

Conclusion

Our results show that the Bioruptor® NGS is an ideal device for mid- to high-throughput NGS labs as it produces reliable and reproducible results in all library preparation protocols that need DNA fragmentation.

* The Bioruptor® NGS is no longer available for sale from Diagenode. The new Bioruptor® Pico is the modern evolution of the Bioruptor® NGS.

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