New automated systems for size-selection in NGS library construction

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Introduction

Virtually all genomic library construction methods use some kind of size-selection in order to avoid adapter artifacts, and to place boundaries on the library fragment size. The gold standard method for size-selection is preparative manual agarose gel electrophoresis, a procedure that is laborious, irreproducible, and difficult to automate. In response to this need, Sage Science introduced its Pippin Prep automated preparative electrophoresis system in 2010. Sage is introducing two new preparative electrophoresis systems. The first system, called the ELF (Electrophoretic Lateral Fractionator) is designed to fractionate a single genomic sample into 12 contiguous size fractions. The system uses a two-dimensional process to separate DNA through an agarose column in a first direction, and then move the separated DNA fragments sideways into a linear array of buffer-filled elution modules that are positioned alongside the separation column. Each disposable cassette will process a single sample, and the instrument will process two cassettes per run. The instrument can operate in direct current mode for samples up to mid-single kilobases in size, or in pulsed field mode for samples up to 50 kilobases in size. Fractionation range is controlled by selection of voltage protocol, gel concentration, and run timing. Internal standards can be used to improve run-to-run reproducibility, and the instrument can read fluorescein-labeled markers.

Examples of ELF performance

2% agarose gel, continuous field, sample of Ecoli genomic DNA digested with 4base cutters

0.75% agarose gel, pulsed field prog. 1, restricted Ecoli gDNA, 2.5 hours

0.75% agarose gel, pulsed field prog. 1, restricted Ecoli gDNA, 5 hours

0.75% agarose gel, pulsed field prog. 2, restricted Ecoli genome DNA, 6 hours

ELF Concept

Step 1. Separation electrophoresis through agarose channel

Step 2. Elution electrophoresis into buffer filled elution module strip

The ELF system is useful for constructing multiple libraries with different insert sizes from the same sample (mate-pair libraries for analysis of structural variation, RNA-seq libraries). The system is also useful for situations when the user wants to recover and save unused portions of the sample for future analyses.

ELF Cassette

ELF Instrument

The ELF instrument will run 2 cassettes (1 sample/cassette). The electrodes are contained in the instrument lid, and are exposed when the lid is closed. The instrument has an on-board computer, and is supplied with a monitor and keyboard. Dimensions: 9.9”(high), 13.3”(wide), 11.5”(deep).

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Coming in Q1-2014: Pippin HT

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