

Purification of Ultra-High Molecular Weight DNA from White Blood Cells on the SageHLS™ System

Introduction

The SageHLS is a new automated platform from Sage Science designed to help scientists work with high molecular weight (HMW) DNA for applications ranging from long-read and long-range sequencing to optical mapping and more. The instrument uses electrophoresis to quiescently purify DNA from cell suspensions, causing intact DNA to become immobilized on an agarose gel surface. The DNA is then re-mobilized by enzymatic cleaving, and collected in six size-fractions using automated preparative electrophoresis.

DNA Purification from White Blood Cells

Goat white blood cells were prepared from whole blood using standard centrifugation techniques. The cells ($\sim 6.6 \times 10^4$) were resuspended in 70 μ l and loaded onto a sageHLS gel cassette. The purification and cleaving processes include several pipetting, electrophoresis, and incubation steps requiring about 90 minutes. Size selection is automated and required 3.5 hours.

Conclusions

Electrophoresis can be used to purify DNA with minimal disruption to its integrity. By adjusting reagent formulations, electrophoresis conditions and temperature, this process can be tuned to meet high molecular weight sample requirements for various optical mapping or long-range sequencing techniques. By providing a temperature-controlled reaction environment, other enzymatic treatments of purified DNA can be utilized prior to collection of size-selected fractions. This could include library construction or CRISPR/Cas9-mediated purification of regions of interest.



sageHLS™ HMW Library System

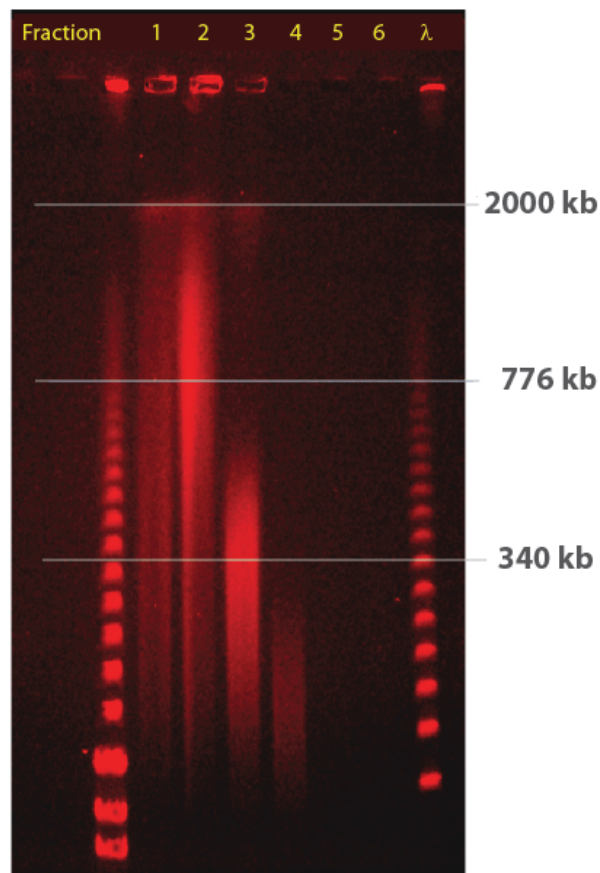


Figure 1. A pulsed-field gel showing purified DNA from white blood cells collected in six size-fraction wells.

Input DNA: 8 μ g DNA equivalents

Fraction	DNA (ng)	% yield
1	428	5.3%
2	1581	19.8%
3	1691	21.1%
4	314	3.9%
5	24	0.3%
6	26	0.3%
Total	4063	51%

Table 1. The relative sample recoveries of the DNA size fractions. DNA amounts were measures using Qubit™ picogreen fluorometric assays.