

Overview

An ideal fragment library would be generated from low amounts of input DNA and would be bias-free. Lucigen is developing a PCR-free, low input fragment library preparation kit to enable the use of low input amounts without the need for PCR amplification prior to sequencing.

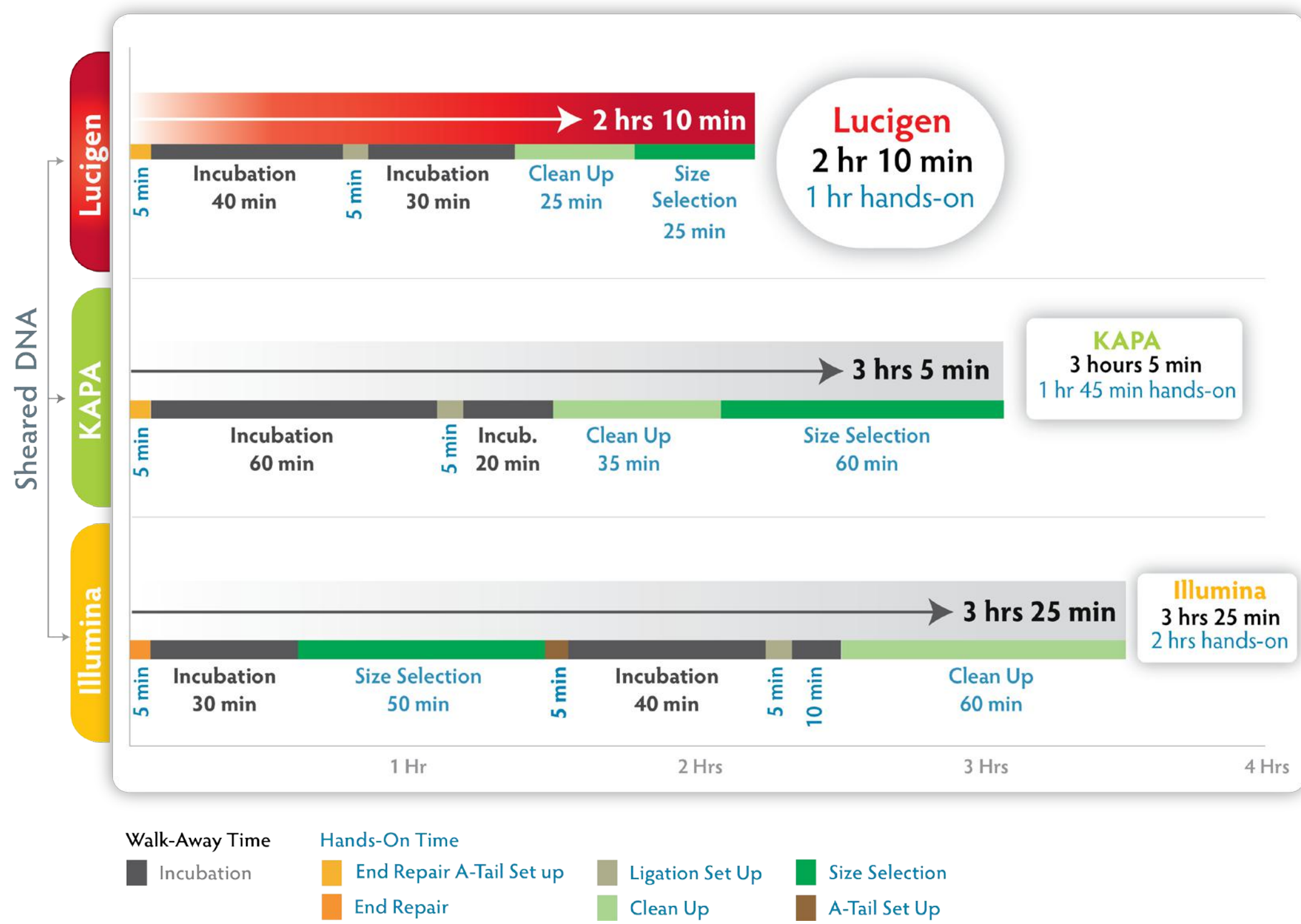
Superior Workflow

Generate Fragment Libraries in 2 hours

Lucigen's automation friendly workflow can be completed in 2 hours with only one clean up and one size selection step.

The Lucigen system requires less time than alternate systems:

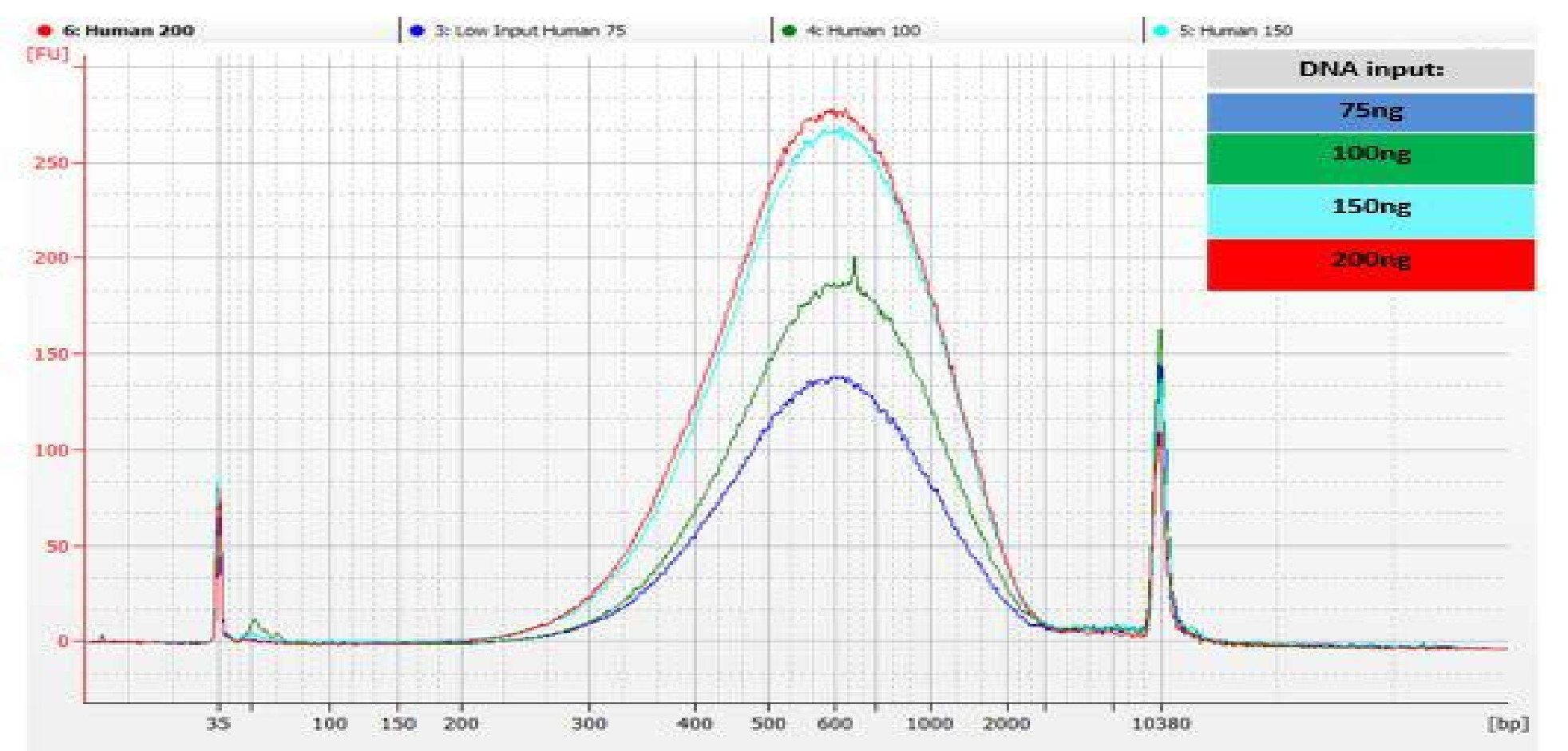
- Illumina TruSeq PCR-free sample prep (cat. #FC-121-3001)
- KAPA Hyper Prep Kit (cat #KK8501)



Conclusions

- Generate fragment libraries in 2 hours
- Generate fragment libraries using 75 ng of input
- No need for amplification prior to sequencing
- Over 95% of reads map to reference genome
- Low GC bias
- This kit launched as the NxSeq® AmpFREE Low DNA Library Kit (Cat. # 14000-1 and -2)

Results



Sequence Without Amplification

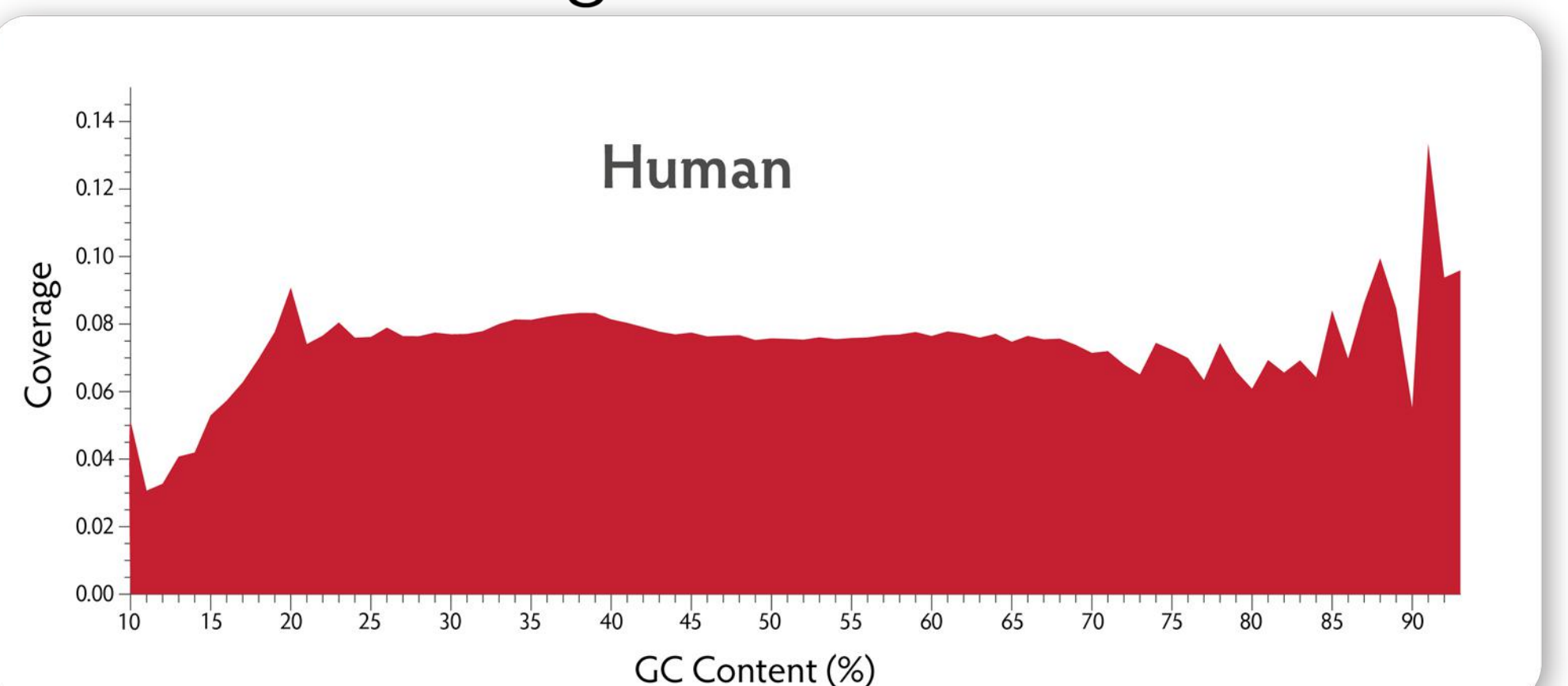
Sufficient library for sequencing is generated using as low as 75 ng input material.

| | Human Library 75 ng input Amount |
|-----------------|-------------------------------------|
| Genome size, GC | ~3 Gbp 45% GC |
| Raw reads | 3,131,114* |
| Mapped reads | 2,979,237 (95.15%) |
| Read length | 148.9 bp |
| Total bases | 443,767,447 |
| Genome fraction | 0.11 |
| Avg. coverage | 0.15X |

Over 95% of Reads Map to Reference Genome

*Library sequencing on a MiSeq using 2x150 bp. Pooled with 2 other libraries prior to sequencing.

Coverage vs GC Content Plot



Low GC Bias

Minimal bias observed when the coverage is plotted against the GC content. The variation at high GC content is expected due to the low frequency of high GC in the human genome.