

KAPA EvoPlus Kits Guide to Success



Streamlined sample prep workflow using the next generation of KAPA DNA Library Prep Reagents.



How much DNA do I need?

Application	Sample Type	Input
WGS	High quality gDNA	10 - 500 ng
WGS (PCR-free)	High quality gDNA	≥ 75 ng (no SS)* 500 ng (w/SS)*
WES**	High quality gDNA	100 ng
	Low quality FFPET derived DNA	> 100 ng

* SS = double-sided size selection: a requirement when performing WGS on patterned flow cells but may result in sample losses of 60 - 95%, irrespective of whether a bead- or gel-based technique is used.
** For this application, please refer to the KAPA HyperCap Workflow v3.x for guidelines.

Get to chopping.

- Mode and size distribution of DNA is controlled by fragmentation time and temperature.
- Try a range of fragmentation times to determine optimal insert size.
- For ease of sample processing, place samples with the longest fragmentation time in the thermal cycler first. Add samples with shorter fragmentation times at appropriate intervals.

Estimated fragment length*	Fragmentation time at 37°C
550 - 640 bp	5 min
330 - 410 bp	10 min
240 - 320 bp	15 min
200 - 260 bp	20 min
170 - 230 bp	25 min
140 - 210 bp	30 min

* Sizes observed upon fragmentation of NA12878 (Coriell DNA). Size variation may be observed, depending on DNA type, DNA input and DNA elution buffers. We recommend optimizing the fragmentation time with a non-precious sample.

How much adapter do I need?
Adapter concentration affects ligation efficiency, as well as adapter and adapter-dimer carry-over during the post-ligation cleanup.

Input DNA	Adapter stock concentration	Adapter: insert molar ratio*
500 ng	15 μM	20:1
100 ng	15 μM	100:1
75 ng	15 μM	135:1
50 ng	15 μM	200:1
10 ng	6 μM	400:1

* Adapter: insert molar ratio calculations are based on a mode DNA fragment length of 200 bp, and will be higher for longer DNA fragments, or slightly lower for DNA fragmented to a mode size <200 bp. The lower adapter: insert molar ratios recommended for inputs >100 ng represent a fair compromise between library construction efficiency and cost; higher library yields will be achieved if a higher adapter concentration is used.

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Evolving together





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