

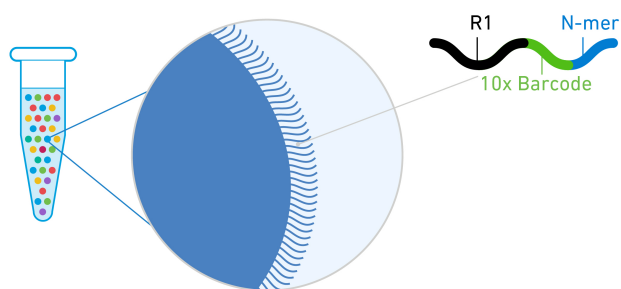
## TECHNICAL NOTE

# Assay Scheme and Configuration of Chromium™ Genome v2 Libraries

### INTRODUCTION

The Chromium™ Genome v2 Protocol (CG00043) produces Genome v2 libraries ready for Illumina® sequencing. During library preparation sequence components essential for Illumina sequencing and downstream data analysis are incorporated into the final library construct. The sequence components are introduced via the 10x™ Gel Beads and the library preparation steps of the workflow.

Each Gel Bead contains millions of oligo primers that are comprised of the following sequences (Figure 1):



- i. Partial Illumina Read 1 sequence (22 nucleotides (nt))
- ii. 16 nt 10x™ Barcode
- iii. 6 nt random N-mer (hexamer)

Fig. 1. Schematic of a Genome v2 Gel Bead oligo primer.

10x Genomics technology is based on the partitioning of samples and reagents into droplets, each called a Gel Bead in Emulsion (GEM). Once partitioned, the Gel Bead dissolves and its oligo primers are released into the aqueous environment of the GEM. The contents of the GEM (oligos, high molecular weight gDNA and Master Mix) are incubated in an isothermal reaction. Random priming by the Gel Bead oligo via the hexamer end and a separate random hexamer supplied in the Master Mix produces barcoded fragments ranging from a few to several hundred base pairs. The GEMs are then “broken”, pooling the barcoded DNA molecules from the separate partitions. During library construction Read 2 is added during Adapter ligation. Illumina P5 and P7 sequences and i7 sample indices are added during Sample Index PCR. The final library fragments contain P5, P7, Read 1 and Read 2 sequences used in Illumina bridge amplification and sequencing. Additionally, each fragment contains a 10x Barcode and the genomic insert (Figure 2). An overview of the Genome v2 assay scheme and how individual sequence components are incorporated during library construction is presented in Figure 3.

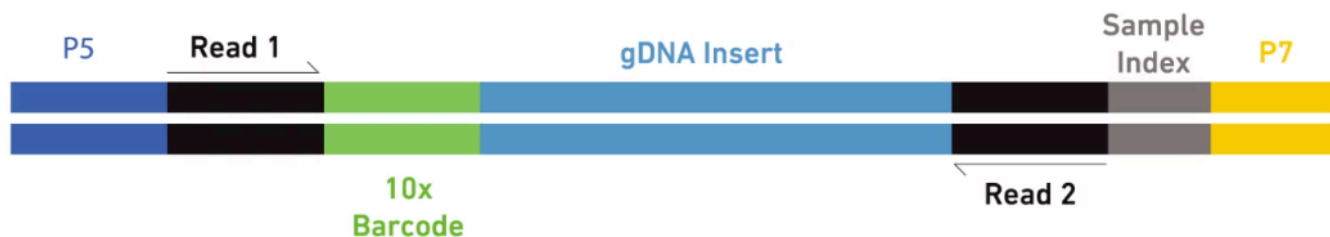


Fig. 2. Schematic of a fragment from a final Chromium™ Genome v2 library.

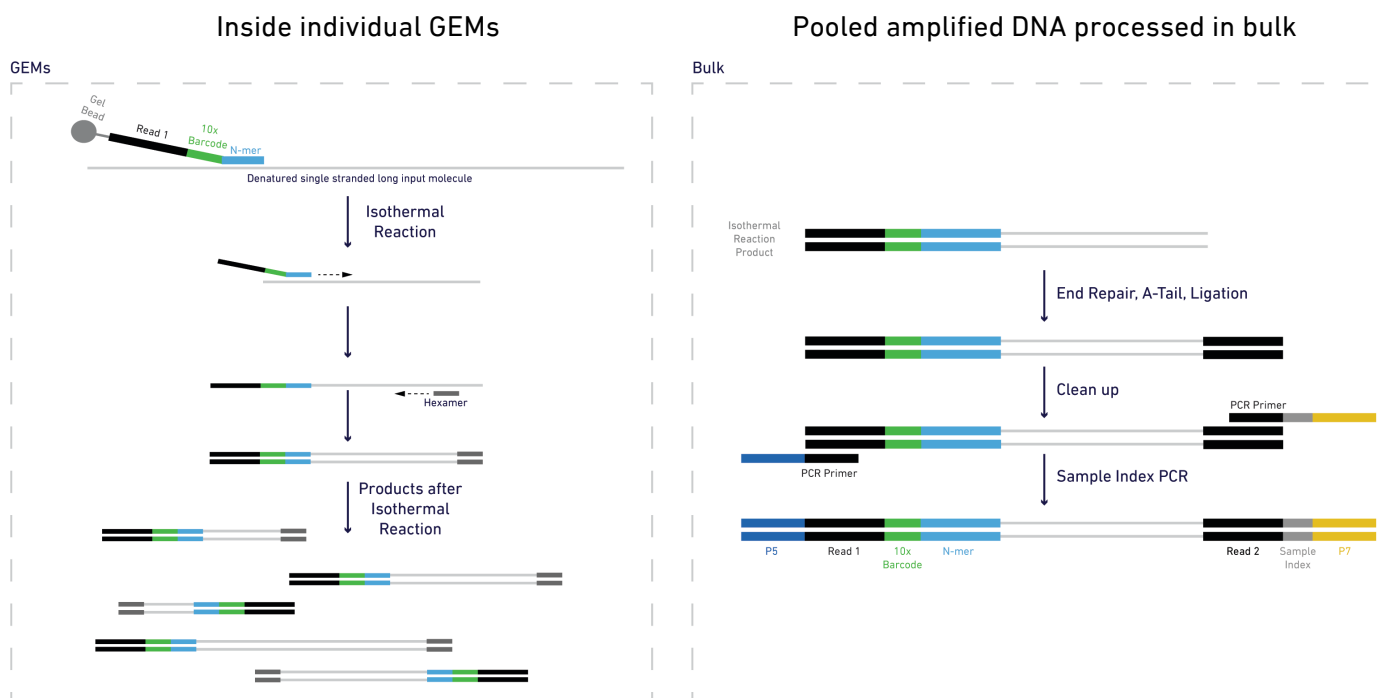


Fig. 3. Schematic of assay scheme for Chromium™ Genome v2 library preparation. The Gel Bead oligo primer randomly binds to multiple locations of HMW gDNA fragments to produce many barcoded products. The binding of only one Gel Bead oligo primer is shown for simplicity.

Figure 4 provides a detailed description of the library preparation workflow. Individual protocol steps that are listed in Figure 4 refer to the *Chromium™ Genome Reagent Kits v2 User Guide* (CG00043).

## CONCLUSION

We have presented a detailed description of the assay configuration for Chromium™ Genome v2 libraries. Individual steps during library construction outlined here provide additional insight and may serve as a reference to customize the library preparation workflow.

## REFERENCES

- *Chromium™ Genome Reagent Kits v2 User Guide* (CG00043)

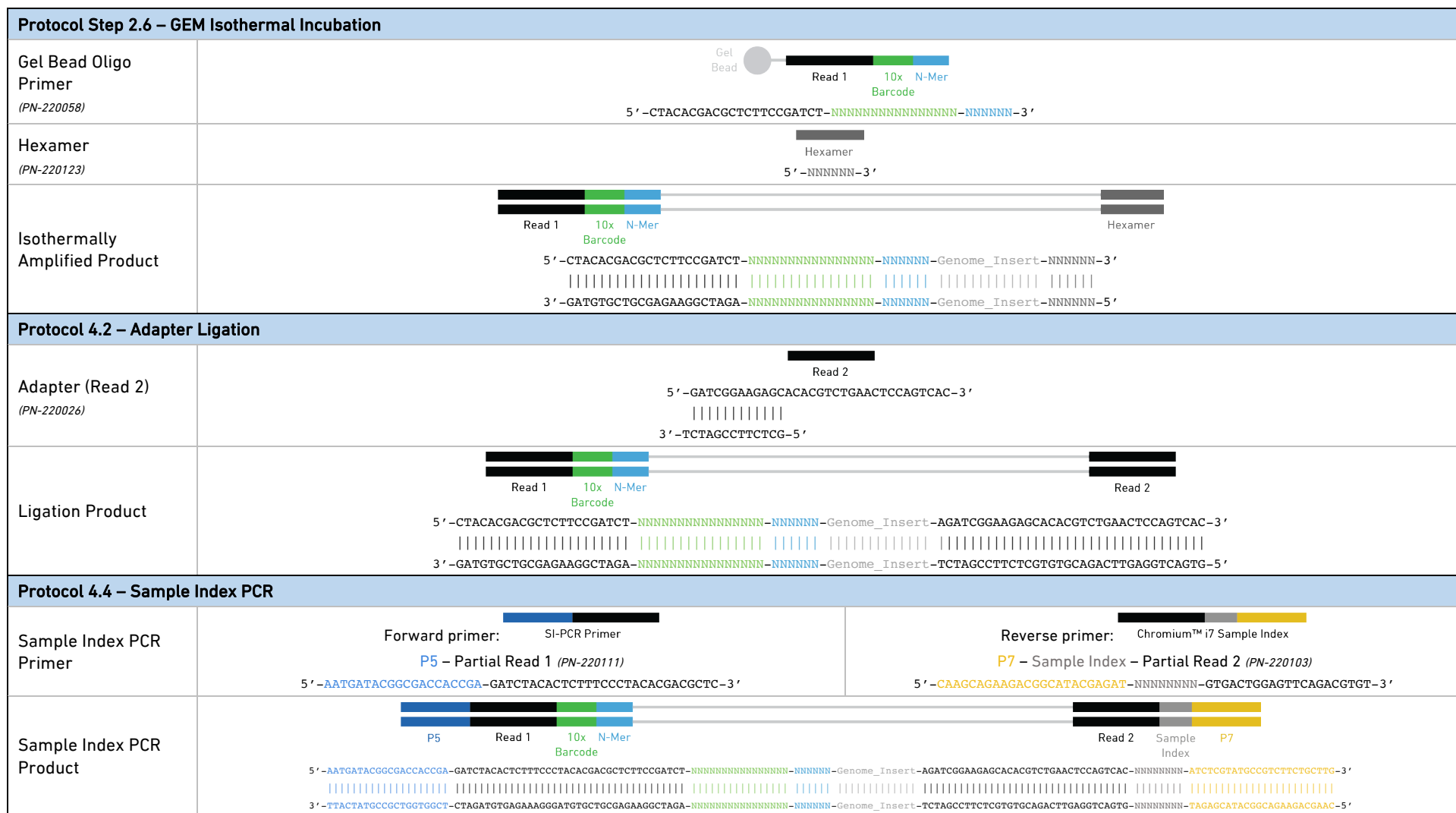


Fig. 4. Flow chart that outlines construction of Single Genome v2 libraries. Protocol steps and part numbers refer to *Chromium™ Genome Reagent Kits v2 User Guide* (CG00043).

# Notices

## Document Number

CG000110 Rev A *Technical Note*

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