

Infinium® Assay Workflow

The Infinium assay provides extensive levels of multiplexing for genome-wide and targeted genotyping applications with a manual or automated workflow.

Introduction

The Infinium Whole-Genome Genotyping assay (Figure 1) is designed to interrogate many single nucleotide polymorphisms (SNPs) at extensive levels of loci multiplexing. Using a single bead type and dual-color channel approach, the Infinium assay scales genotyping from hundreds to millions of SNPs per sample. The optional Illumina Laboratory Information Management System (LIMS) and automation with a Tecan liquid-handling robot help ensure positive sample tracking while reducing hands-on time and labor costs.

Infinium Assay Workflow

The Infinium assay features a relatively low input DNA sample requirement of 200 ng, which is sufficient to assay millions of SNP loci. The Infinium assay workflow (Figure 1) begins on day one with an overnight amplification of the DNA sample (Step 1). This amplification has no appreciable allelic partiality. On day two the amplified product undergoes controlled enzymatic fragmentation, which does not require gel electrophoresis (Step 2). After alcohol precipitation and DNA resuspension (Step 3), the BeadChip is prepared for hybridization in the capillary flow-through chamber (Step 4). Samples are applied to prepared BeadChips and incubated overnight. During this overnight hybridization, the DNA samples anneal to locus-specific 50-mers covalently linked to up to millions of bead types (Step 5). One bead type corresponds to each allele per SNP locus. On day three, allelic specificity is conferred by enzymatic base extension followed by fluorescent staining (Step 6). The iScan® System detects the fluorescence intensities of the beads (Step 7),

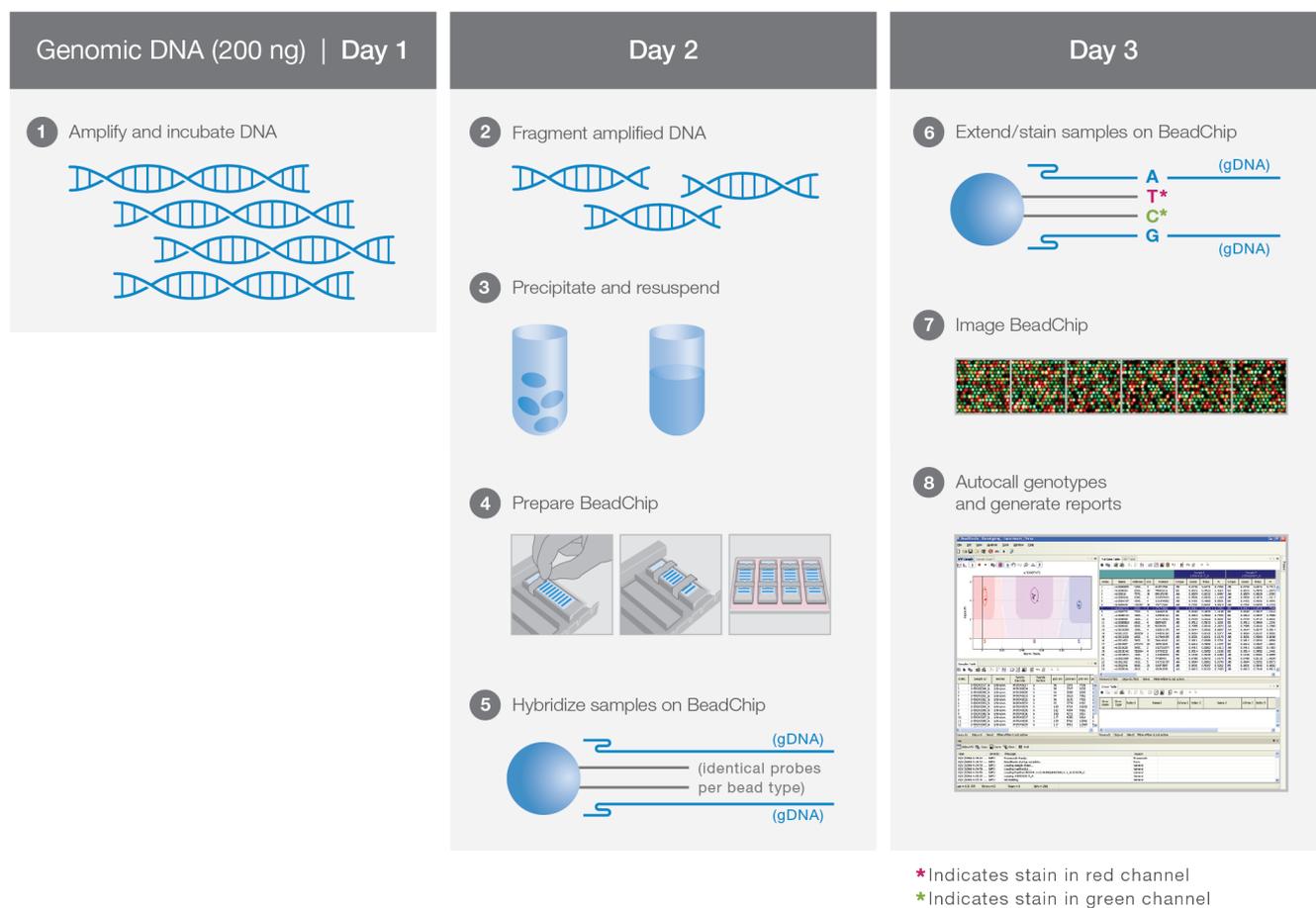


Figure 1: The Infinium Assay Workflow—The Infinium assay workflow proceeds from input DNA to automated genotype report with a total assay turnaround time of three days.

Table 1: Infinium Assay Workflow Using the Global Screening Array-24 v1.0 BeadChip^a

Day	Step	Automated Workflow (Robot Time and Hands-on Time) ^b	Manual Workflow (Hands-on Time) ^b	Incubation/Dry Time ^b
1	Set up DNA Amplification	1 hr (robot) 15 min (hands-on)	1 hr	20–24 hr
	Fragment Amplified DNA	10 min (robot) 5 min (hands-on)	30 min	1 hr
2	Precipitate Amplified DNA	20 min (robot) 5 min (hands-on)	30 min	2 hr
	Resuspend Amplified DNA	15 min (robot) 5 min (hands-on)	30 min	1 hr
	Hybridize Sample to BeadChip	25 min (robot) 20 min (hands-on)	20 min	16–24 hr
3	Wash BeadChip	20 min (hands-on)	20 min	25 min
	Stain BeadChip	2 hr 45 min (robot) 5 min (hands-on)	2 hr 45 min	1 hr
	Image BeadChip	2 hr (robot) 5 min (hands-on)	2 hr (robot) 5 min (hands-on)	—

a. Times indicated for processing 4 BeadChips and 96 samples using the iScan System.

b. Times listed are approximate, actual times may vary.

and Illumina software automatically performs analysis and genotype calling (Step 8). The [Infinium XT Production-Scale Genotyping Solution](#) features an optimized version of the Infinium assay workflow with user-friendly enhancements and reductions in total assay turnaround time and hands-on time.

Estimated processing times required for completing the Infinium assay, for both automated and manual workflows, using the Infinium Global Screening Array-24 v1.0 BeadChip and the iScan System are provided (Table 1).

Learn More

To learn more about Infinium products and solutions, visit www.illumina.com/techniques/microarrays.html.