

Illumina custom microarray (EPIC+)



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Array Design

Microarray technology has provided a cost-effective platform for various DNA methylation studies. Existing DNA methylation arrays have been robustly developed for high reproducibility and comprehensive coverage of genomic sites. We have developed a custom DNA methylation microarray that relies on the same robust technology to produce results for an even greater number of genomic sites.

The methylation array utilized here is a custom Illumina® Infinium™ BeadChip designed to have more comprehensive coverage of the human genome than the existing industry standard manufactured by Illumina. The base for this custom array is the Infinium MethylationEPIC v1.0 B4 (EPIC) BeadChip, Illumina's current, most comprehensive array. The custom array includes all EPIC probes with the addition of 2,570 probes that were previously included in the Infinium™ HumanMethylation450K (450K) BeadChip, but excluded from the EPIC BeadChip. These probes from the 450K array were selected based on evidence among validated methylome-wide association studies, as well as human population studies. Specific probes of interest were captured in replicate to ensure quantification by the array.

Increased Genome-Wide Coverage

The methylation array interrogates a resulting set of 868,488 unique sites, 2,570 more than the EPIC array, and 382,911 more than the 450K array. These sites include 865,492 CpGs, 2,932 non-CpG (probes with the prefix "ch"), and 64 SNPs (Table 1).

Table 1. Number of each site type covered by probes in three DNA methylation arrays. Numbers are presented as n (%).

Site type	Custom array	EPIC array	450k array
CpG	865,492 (99.6%)	862,927 (99.6%)	482,421 (99.3%)
Non-CpG	2,932 (0.3%)	2,932 (0.3%)	3,091 (0.6%)
SNP	64 (0.1%)	59 (0.1%)	65 (0.1%)

The custom methylation array takes advantage of, and expands upon, the already vast coverage of the methylome offered by existing arrays. Specifically, there is increased coverage of CpG islands and CpG island regions (Table 2). Furthermore, the array includes probes that span multiple gene regions. Table 3 outlines the specific types of UCSC RefGene features covered by the array.

Table 2. CpG island coverage of probes on DNA methylation microarrays.

Array	N Shelf	N Shore	CpG Island	S Shore	S Shelf
Custom	32,118	83,792	162,237	71,490	29,797
EPIC	31,991	83,364	161,441	71,182	29,700
450k	24,844	62,870	150,254	49,197	22,300

Table 3. UCSC RefGene feature coverage of probes on DNA methylation microarrays.

Site type	Custom array	EPIC array	450k array
TSS1500	201,465	200,780	122,745
TSS200	122,086	121,655	89,029
5'UTR	201,611	200,962	103,241
1stExon	70,642	70,366	53,840
Body	711,248	70,9569	288,345
3'UTR	39,712	39,553	29,937

Reproducibility

The custom array was evaluated for reproducibility based on the correlation of results of a well-characterized, human male sample (NA10858) from the Coriell Institute for Medical Research (Camden, NJ). The custom methylation array exhibits a strong correlation with existing Illumina BeadChip arrays (Pearson R = 0.9824, $p < 0.0001$, Figure 1). Additionally, the custom array shows high reproducibility between technical replicates (Pearson R = 0.9975, $p < 0.0001$, Figure 2).

Figure 1

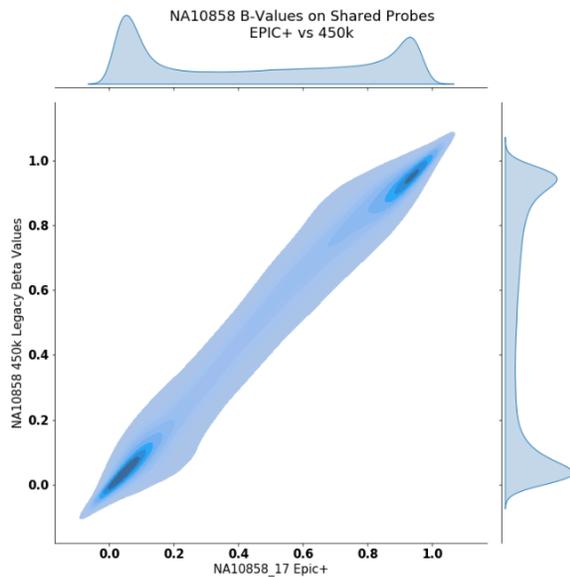


Figure 2

