There are 850 inbuilt control probes on the 450K array, such as bisulfite conversion I, bisulfite conversion II, extension, hybridization and negative (n = 613), which can be used to evaluate the other probes’ intensities. Samples that can not pass this quality control are excluded in further analysis.

485512+850+65=486427

Table 1. Control Probes on the Illumina 450K methylation array

|  |  |  |  |
| --- | --- | --- | --- |
|  | Description | No. of Types | No. of Probes |
| Bisulfite Conversion | Methylation at a site known to be methylated | 3 | 16 |
| Normalisation | Randomly permutated bisulphite-converted sequences containing no CpGs; Determines system background (A,T,C,G, 32, 61, 61,32) | 4 | 186 |
| Staining | Efficiency and sensitivity of staining step | 2 | 6 |
| Extension | Extension efficiency of A, T, C, and G nucleotides from a hairpin probe | 4 | 4 |
| Hybridisation | Hybridisation efficiency using synthetic targets instead of amplified DNA | 3 | 3 |
| Target Removal | Efficiency of stripping step after extension reaction | 1 | 2 |
| Specificity | Methylation at non-polymorphic T sites | 3 | 16 |
| Non-polymorphic | Methylation at a base in a non-polymorphic region of the genome | 4 | 4 |
| Negative |  |  | 613 |
|  |  |  | Total: 850 |
| SNP1 |  |  | 25 |
| SNPII |  |  | 40 |
|  |  |  |  |