MaizeSNP50 BeadChip

Enabling the most comprehensive interrogation of maize genetic variation.

MaizeSNP50 BeadChip Highlights

- **Unrivaled Call Rates and Accuracy**
  > 99% average call rates and 99.99% reproducibility
- **Comprehensive and Uniform Coverage**
  Genome-wide coverage using over 55,000 evenly spaced markers
- **Simple Workflow**
  PCR-free protocol with the powerful Infinium® HD Assay
- **High-Throughput Format**
  Up to 24 samples interrogated in parallel

Introduction

Maize (*Zea mays* L. ssp. *mays*) was domesticated from the grass teosinte in Central America[1] over the last ~10,000 years. It is an important model organism for fundamental research into the inheritance and functions of genes[2], as well as an important crop. In 2008, maize cultivation in the United States yielded 12 billion bushels of grain from approximately 86 million acres with a value of $47 billion[3].

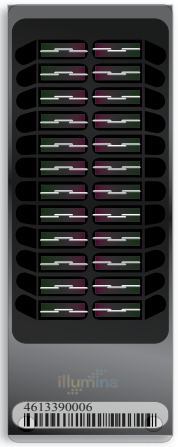
The maize genome has undergone several rounds of genome duplication. Its 10 chromosomes are structurally diverse and have endured dynamic changes in chromatin composition. Over the last three million years, the size of the maize genome has expanded dramatically (to 2.3 gigabases) via a proliferation of long terminal repeat (LTR) retrotransposons[4].

Maize exhibits extremely high levels of both phenotypic and genetic diversity. It has been suggested that the diversity between maize lines is greater than the diversity between humans and chimpanzees.

In 2009, researchers completed an improved draft nucleotide sequence of the 2.3-gigabase genome of the B73 maize line[5]. This assembly was used to compare sequence data of other key maize lines. From these efforts over one million SNPs have been identified[6].

The MaizeSNP50 Genotyping BeadChip contains 56,110 markers derived from the B73 reference sequence (Figure 1). Illumina has developed the BeadChip in collaboration with TraitGenetics, The French National Institute for Agricultural Research (INRA), and Syngenta. Featuring highly polymorphic SNP content and providing uniform genomic coverage (Figure 2), this BeadChip enables the interrogation of genetic variation for maize. Importantly, this BeadChip presents an average of greater than 25 markers per megabase (Mb), providing ample SNP density for robust whole-genome genotyping studies. In addition, the MaizeSNP50 marker set provides the ability to perform genetic mapping and marker-assisted breeding. This research will facilitate efforts to meet the world's growing needs for food, feed, energy, and industrial feed stocks in an era of global climate change.

Figure 1: MaizeSNP50 BeadChip



The MaizeSNP50 Genotyping BeadChip features more than 55,000 evenly spaced SNPs across the entire maize genome.

The combination of Illumina's proprietary assay technologies, unconstrained locus selection, and high-throughput format presents the most comprehensive solution for agrigenomic studies in maize.

SNP Selection Strategy

The SNP content featured on the MaizeSNP50 BeadChip was selected from several public[6] and private sources. Considerations for SNP selection included genic representation, optimized physical spacing, and diversity line identification for multiple known lines. This content selection strategy empowers confident genetic mapping in maize, demonstrated by the successful validation in more than 30 diverse lines (Table 1).

High-Quality Data

The MaizeSNP50 BeadChip is powered by the proven Infinium HD Assay, providing the industry's highest call rates and flexible content design. SNPs on the MaizeSNP50 were subjected to rigorous functional testing across multiple lines to ensure strong performance.

Because of the diversity of the maize genome, some sequences are not present in all lines. This can result in lower call frequencies for lines that diverge from the B73 reference genome. The approximately 4,000 markers in the product performed well on B73 samples, but not as well on samples that differ from B73. These markers have been identified and are provided in annotation files. Similarly, a subset of approximately 500 markers performed suboptimally on samples derived from B73, but well on other samples.

AGAATGATAACAGTAACACACTTCTGTTAACCTTAAGATTACTTGATCCACTGATTCAACGTACCGTAACGAACGTATCAATTGAGACTAAATATTAACGTACCATTAAGAGCTACCGTCTTCTGTTAACCTTAAGATTACTTGATCCACTGATTCAAC
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CGTATCAATTGAGACTAAATATTAACGTACCATTAAGAGTCTGTTAACCTTAAGATTACTTGATCCACTGATTCAACGTACCGTAACGAACGTATCAATTGAGACTAAATATTAACGTACCATTAAGAGCTACCGTGCAACGAAAAGAATGATAACAGTA

Table 1: Diversity of Lines Used to Evaluate the MaizeSNP50 BeadChip

Lines	Number of DNA Samples	Call Rate (%)	Heterozygosity Rate (%)
B73	3	99.40	0.32
Mo17	3	97.37	2.50
B73xMO17	3	97.34	51.84
25 DL*	25	96.34	2.26
NAM F1s†	25	97.61	40.89
IBMs‡	94	97.59	2.17
PVPs§	96	95.35	2.77
Teosinte	6	91.67	2.52
TeoNIL F1	6	88.10	34.02
LHRE¶	94	95.08	4.29
All	489	95.65	6.86

* DL = Diverse Lines
† NAM = Nested Association Mapping population
‡ IBM = Intermediated B73 x Mo17 mapping population
§ PVP = Plant Variety Protection Act
¶ LHRE = Lines Highly Recombinant European

Illumina's internal validation of the MaizeSNP50 BeadChip is shown in Table 2. For samples representing the B73 reference genome line, call rates across 56,110 markers were 99.40% with high reproducibility and low Mendelian inconsistency rates. Across 489 samples, spanning more than 30 diverse maize lines, the call rate was 95.65%. Reproducibility was > 99.9% for nine replicates and the Mendelian inconsistency was 0.16% for 43 inbred crosses.

Figure 2 shows the distribution of the markers for the MaizeSNP50 BeadChip. More than 50% of the markers are within 10,000 nucleotides of the neighboring marker. Less than 5% of the physical gaps are larger than 200 Kb. With more than 55,000 markers, the average spacing is one marker every 40 Kb.

Call rates and heterozygosity for the diversity lines used to validate markers for the MaizeSNP50 BeadChip are shown in Table 1. The three B73 reference samples have an average call rate of > 99%, while the six Teosinte progenitor samples have an average call rate of 91%. Samples evaluated include a panel of 25 highly diverse inbred lines, as well as their crosses with B73. Importantly, the inbred cross of B73 x Mo17 is heterozygous for more than 50% of markers contained in the panel. Samples for marker validation also included European and North American mapping populations, as well as the Plant Variety Protection Act panel.[7]

Illumina Solutions for Genotyping

The high-quality data and low per-sample cost of the MaizeSNP50 BeadChip are part of the powerful Illumina Whole-Genome Genotyping Solution. In addition, optional automation and the Laboratory Information Management System (LIMS) lower costs by eliminating errors associated with manual processing. Illumina genotyping products can be accessed via Illumina FastTrack Genotyping Services or any Illumina CSPro® Service. Illumina solutions provide industry-leading levels of accuracy, flexibility, and affordability. To learn more about Illumina's genotyping solutions, please visit www.illumina.com/maizeSNP50.

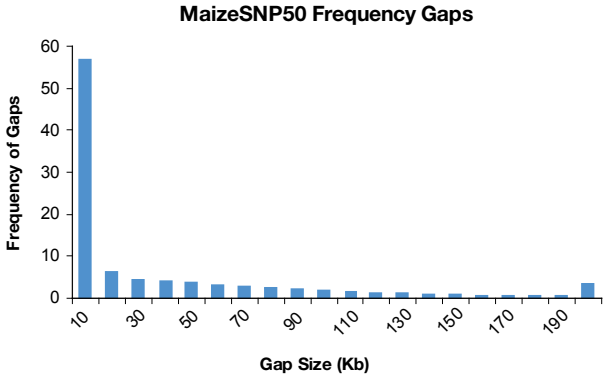
Table 2: MaizeSNP50 BeadChip Performance Data and Specifications

Parameter	Percent	Product Specification
Average Call Rate	99.40%*	> 99%
Reproducibility	> 99.9%	> 99.9%
Mendelian Inconsistencies	0.036%	< 0.1%

*Maize line B73 only

A standard cluster file is provided with the product to use as a starting point for calling genotypes. Given the complexity and diversity of maize lines, it is important to evaluate cluster positions for each data set. It is also best practice to include parental control inbred lines and their crosses for reference.

Figure 2: MaizeSNP50 BeadChip Probe Spacing



The MaizeSNP50 BeadChip provides uniform coverage across the entire maize genome.

References

1. Doebley JF et al. (2006) The molecular genetics of crop domestication, Cell 127:1309–21.
2. J. L. Bennetzen, S. Hake, Handbook of Maize: Genetics and Genomics (Springer, New York, 2009).
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Ordering Information

Catalog No.	Product	Description
WG-500-1001	MaizeSNP50 Whole-Genome Genotyping Kit (48 samples)	Each package contains 2 BeadChips and reagents for processing 48 samples.
WG-500-1002	MaizeSNP50 Whole-Genome Genotyping Kit (288 samples)	Each package contains 12 BeadChips and reagents for processing 288 samples.
WG-500-1003	MaizeSNP50 Whole-Genome Genotyping Kit (1152 samples)	Each package contains 48 BeadChips and reagents for processing 1152 samples.
FT-440-1006	MaizeSNP50 FastTrack Service Project	Illumina's FastTrack provides services for a wide range of SNP genotyping projects.

Each MaizeSNP50 Genotyping BeadChip can process 24 samples in parallel and assay 56,110 markers per sample.

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