

Axiom® Strawberry Genotyping Array

The world's first expert-designed octoploid strawberry genotyping array to facilitate Quantitative Trait Loci (QTL) discovery and marker-assisted breeding

Axiom® Strawberry Genotyping Array (also called the International Strawberry 90K (IStraw90) Axiom® Array), was designed through Affymetrix® Expert Design Program in collaboration with the International RosBREED SNP consortium that spans multiple nations, states, and institutions dedicated to genetic improvement of rosaceous crops.

The array offers genome-wide coverage of polymorphic SNPs across the cultivated garden strawberry hybrid (*Fragaria* × *ananassa*).

Highlights

- **Diverse content:** 95,062 SNPs and indels derived from octoploid and diploid cultivars, including:
 - 1,761 multi-allelic SNPs
 - 3,751 SNPs derived from diploid cultivars; the remaining markers are derived from octoploid cultivars
- **Multiple cultivars represented:** SNP discovery was facilitated with a diverse worldwide breeding germplasm panel

Applications

- **Complex trait research:**
 - QTL discovery
 - Identify traits of economic significance
- **Molecular breeding:**
 - Genome-wide scanning
 - Marker-assisted breeding
- **Release of new cultivars:**
 - Accelerate and increase efficiency of cultivar development
 - Identify the presence of important markers in existing strawberry germplasm
 - Use genetic information to identify germplasm that will perform well in specific growing regions

SNP discovery

SNP discovery was facilitated by sequencing a diverse discovery panel of 9 octoploid cultivars at ≥20x coverage, including:

- Holiday, Korona, and F1 seedlings from the cross Holiday × Korona.
- Two likely diploid progenitors, *Fragaria mandschurica*, *F. iinumae*
- One known diploid, *F. vesca*

The sequencing reads were aligned to the *F. vesca* genome sequence.

Marker selection

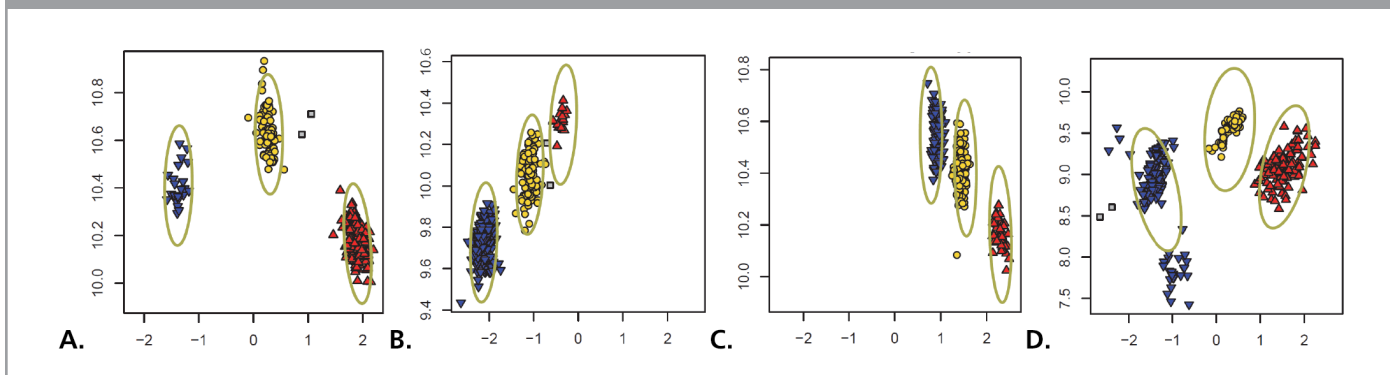
Markers were selected with the goal of addressing the challenges in genotyping the allo-octoploid strawberry genome to maximize the number of functional markers. These goals were achieved by minimizing sequencing errors and exploiting site-specific biological reductions in ploidy. The SNPs and indels were chosen based on the following criteria:

- Select markers from cultivars that were sequenced at 20x coverage to minimize errors from sequencing, misalignments, or erroneous mappings
- Exclude highly repetitive sequences and those containing ambiguities
- Avoid homopolymer and di-nucleotide repeat sites when selecting indels
- Include SNPs specific to 1 subgenome
- Achieve reduction in ploidy by:
 - Targeting subgenome-specific deletions
 - Targeting sites of subgenome-specific sequence motifs
- Include markers that meet the threshold criteria for predicting reproducibility on the array based on Affymetrix' *in silico* design scores

Experimental results

384 samples were genotyped with the Axiom Strawberry Genotyping Array. The data analysis and clustering were automated using Affymetrix® Genotyping Console™ Software as per the *Best Practice Supplement to Axiom® Genotyping Solution Data Analysis* (P/N 703083) and SNPs were filtered using the Affymetrix® SNPolisher™ package.

Figure 1: The cluster plots below show patterns that were observed in the data generated on Axiom® Strawberry Genotyping Array. Plots A–B are the result of the ploidy reduction strategies used during array design: (A) diploid-like pattern (B) tetraploid-like pattern (C) octoploid pattern. Cluster plot (D) shows an example of a multi-cluster polymorphic high-resolution genotype.



15,000 polymorphic high-resolution markers were selected based on cluster properties to successfully place the SNPs on a genetic map of the hybrid Holiday x Korona¹. Examples of the different types of cluster plots generated during the analysis are shown in Figure 1.

Notes about the data analysis: Axiom® Strawberry Genotyping Array requires advanced analysis using functions provided by the Affymetrix SNPolar R package to filter multi-cluster SNPs such as shown in Figure D. Additional filters based on reproducibility and inheritance errors are also key and require genotyping technical replicates and sets of parents and F1

seedlings. Genotyping many different varieties together may create multiple clusters during the analysis that interfere with each other and/or blend the clusters together, compromising the algorithm's ability to correctly call genotypes. Genotype calls are not provided for multi-allelic SNPs.

Reference

1. Bassil, N. Development and Preliminary Evaluation of the IStraw90 Axiom® Array in Cultivated Strawberry (*Fragaria × ananassa*). *American Society for Horticultural Science oral presentation* (2013).

Ordering information

Part number	Product name	Description
550466	Axiom® Strawberry Genotyping Array (IStraw90K)	Contains one plate with 96 arrays. Reagents and GeneTitan® MC consumables must be quoted separately.
901606	Axiom GeneTitan® Consumables Kit	Contains all GeneTitan® Instrument consumables required to process one array plate
901758	Axiom® 2.0 Reagent Kit	Includes all reagents (except isopropanol) for processing 96 DNA samples

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