Axiom Microbiome Array

Easy, streamlined solution for microbial profiling

The Applied Biosystems™ Axiom™ Microbiome Array enables researchers to detect all known microorganisms in a sample with a single assay. Designed in collaboration with the Lawrence Livermore National Laboratory, the Axiom Microbiome Array provides species- and strain-level detection on a scalable platform with straightforward, easy-to-use software.

Using Applied Biosystems™ Axiom™ assay biochemistry, the Axiom Microbiome Array interrogates nonpolymorphic sequences in both family-conserved and target-specific regions from NCBI database sequences. The Axiom Microbiome Array detects over 12,000 species, including archaea, bacteria, fungi, protozoa, and viruses. The array content is sample-type agnostic, suitable for applications in nutrigenomics, agrigenomics, and animal research and modeling.

Highlights

- Comprehensive coverage of archaea, bacteria, fungi, protozoa, and viruses
- Species- and strain-level detection; RNA virus detection using cDNA template
- Scalable platform with easy-to-use analysis software

Applications

Nutrigenomics research (including the influences of microbial communities on nutrition and diet-related health)

- Understanding the linkage of human gut microbiota to disease states (e.g., ulcerative colitis [1], intestinal inflammation [2], and cardiovascular disease [3])
- High-resolution profiling of probiotic and prebiotic mixtures to understand their impact on the human gut microbiome [4,5]

Agrigenomics

- Assessment of animal gut microbial communities for feed optimization [6]
- Evaluation of microbiota for soil productivity [7]
- Ascertainment of livestock animal health [8]

Animal research and modeling

- Microbial profiling for pathogen detection
- Vivarium screening (monitoring for environmental and animal colony health)
- Disease prevention
- Evaluation of microbial status and response to treatment [9]

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Features

The Applied Biosystems™ Axiom™ Microbiome Solution (Figure 1) enables the detection and profiling of microbial targets in a sample with a single assay.

Array content

The Axiom Microbiome Array provides the power to detect microbial content in complex samples across five groups of organisms. The array content includes probes to more than 12,000 species and allows detection to species and strain levels where available. Table 1 provides a detailed list of target categories on the array. The array plates are available in 24- and 96-array formats.

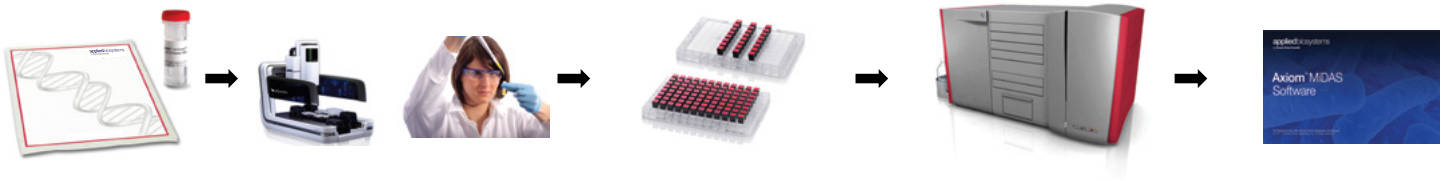
Assay

Utilizing the robust biochemistry of the Axiom 2.0 assay coupled with manual and automated target preparation methods, this streamlined assay protocol provides consistent and high-quality results. A simplified, upstream reverse transcriptase reaction enables the detection of RNA virus genomes. The Applied Biosystems™ GeneTitan™ Multi-Channel (MC) Instrument offers automated processing of Axiom arrays.

Analysis

The analysis workflow requires the use of Applied Biosystems™ Axiom™ Microbial Detection Analysis Software (MiDAS), which is based on the Composite Likelihood Maximization (CLiMax) algorithm [10,11] developed by Lawrence Livermore National Laboratory. The software provides streamlined prediction of target identities in Axiom Microbiome Array data from an unknown sample. Features include complete analysis to strain level, direct access to external NCBI databases, lists of microbial targets most likely present in each sample, and summaries of microbial content in both table and graphical formats to detect and profile the genetic composition of all microbes in a sample.

Axiom MiDAS is available for download from the Axiom MiDAS product web page. The Axiom Microbiome Solution User Guide (P/N 703408) details the workflow required for analysis of the Axiom Microbiome Array.



Axiom 2.0 Reagent Kit
Proven robust and reliable assay

Target preparation
Automated and manual protocols

Predesigned arrays
Available in 24- and 96-array plates

GeneTitan MC Instrument
Automated, hands-free array processing

Axiom MiDAS
Automated microbial profiling

Figure 1. The Axiom Microbiome Solution workflow. From cDNA or genomic DNA to easy-to-use Axiom Microbial Detection Analysis Software (MiDAS), the Axiom Microbiome Solution provides the complete answer for microbial profiling.

Table 1. Target categories represented on the Axiom Microbiome Array.

Category	Number of families*	Number of species	Target sequences**
Archaea	31	370	606
Bacteria	278	6,901	34,254
Fungi	121	381	658
Protozoa	30	91	229
Viral	100	4,770	99,808
Total	**560**	**12,513**	**135,555**

* Number of families reflects NCBI known family classifications as of October 2014. Unknown or ambiguous family-associated targets are not included in the total number of families count, but may be included with "unclassified" or "unknown" family assignments in software results and output files.

** Multiple probes have been designed to interrogate each target sequence. A probe may be common in more than one organism (family-conserved) or unique to a particular strain (target-specific).

Performance

Array performance has been evaluated on both known complex mixtures and bona fide biological samples (stool) against stringent quality control metrics covering positive predictive value (PPV, a measure of precision), true positive rate (TPR, a measure of sensitivity), limit of detection (LOD), and reproducibility. Table 2 contains performance metrics and validation data. TPR and PPV were evaluated on 222 samples of known composition with complexity varying from 1 to 22 strains per sample. A consensus set of expected organisms was identified for all replicates of a sample to evaluate reproducibility (consensus hit rate and consensus precision). Consensus hit rate was calculated as the average percentage of organisms from the consensus set identified by Axiom MiDAS in each replicate. Consensus precision was calculated as the average percentage of organisms detected by Axiom MiDAS in each sample that were present in the consensus set. LOD was determined using a log dilution series of *Thermotoga maritima* genomic DNA (gDNA) by adding 1 to 1,000,000 genome equivalents in the presence or absence of increasing human gDNA. the Axiom Microbiome Array can detect 1,000–10,000 copies of the *T. maritima* genome in the presence of 1–10 ng of human host gDNA with species and strain resolution, and down to 100 copies with genus resolution.

Strain-level resolution is dependent upon sequence information in the reference database. For example, highly related strains or incompletely annotated draft sequences may share probes with the detected strain due to similarity of genomic sequence. This probe sharing can lead to a database target with less complete annotation being the best explanation of the summarized probe intensity data. Axiom MiDAS provides information on alternative targets related to the detected strains, which can be evaluated for further analysis on the strains present in each sample.

Table 2. Axiom Microbiome Array performance metrics.

Metric	Genus	Species	Strain
True positive rate (TPR)	99.1%	96.6%	69.3%
Positive predictive value (PPV)	96.2%	92.6%	61.3%
Limit of detection (LOD)*	100–1,000	1,000–10,000	1,000–10,000
Reproducibility			
Consensus hit rate	98.4%	95.8%	94.0%
Consensus precision	95.3%	94.1%	89.9%

* LOD is reported in genome copies detected.

Acknowledgements

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References

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Ordering information

Product	Description	Cat. No.
Axiom Microbiome 24-Array Plate	Contains one 24-array plate; reagents and GeneTitan Multi-Channel Instrument consumables sold separately	902903
Axiom Microbiome 96-Array Plate	Contains one 96-array plate; reagents and GeneTitan Multi-Channel Instrument consumables sold separately	902904
Axiom Microbiome Reagent Kit for four 24-array plates	Includes all reagents (except isopropanol) to process four 24-array plates	902910
Axiom GeneTitan Consumables Kit	Includes all GeneTitan Multi-Channel Instrument consumables required to process one Axiom 96-array plate	901606
Axiom 2.0 Reagent Kit	Includes all reagents (except isopropanol) to process one 96-array plate	901758

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