DataSheet

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Axiom® Transplant Genotyping Array

Translating transplant genetics into clinical practice to improve transplant success

Genetic research has enabled advancements in human leukocyte antigen (HLA) matching of donor-recipient (D-R) pairs; however, the impact of non-HLA-related genetic variability is not well understood. These non-HLA polymorphisms may be responsible for affecting histocompatibility, allograft rejection, and patient response to immunosuppressive therapies (ISTs).[1] International Genetics & Translational Research in Transplantation Network (iGeneTRAiN) has designed an array tailored for transplantation researchers and immunologists to identify these genetic underpinnings of rejections and complications of transplantation. iGeneTRAiN aims to improve transplantation success through patient-specific IST selection and dosing, enhanced D-R matching, and better monitoring to detect early signs of rejection.

Array highlights

Covering HLA, killer-cell immunoglobulin-like receptor (KIR), pharmacogenomic, and metabolic loci related to transplantation outcomes,[2] Axiom Transplant Genotyping Array enables cost-effective, large-scale genotyping of transplant-related studies. An optimized genome-wide association grid allows for imputation across African, Asian, and European populations, while the functional variants modules provide focused coverage of loss-of-function (LoF), untranslated, and copy number (CN) regions.

Applications

- Transplantation research
- Pharmaceutical applications
- Immunological research
- Solid organ research

Array content[2]

Axiom Transplant Genotyping Array is available with content modules specifically designed for transplantation research and genome-wide coverage optimized for major populations. Axiom Biobank Plus Genotyping Array (custom) allows you to customize this array for your individual study needs by optimizing for any population while adding, removing, or replacing content in any category.

Genome-wide association study (GWAS) markers were selected for major populations as defined by the HapMap project.[3] These include individuals of African (African ancestry in Southwest Texas [ASW] and Yoruba in Ibadan, Nigeria [YRI]); Asian (Chinese in Metropolitan Denver, CO [CHD]); Han Chinese from Beijing, China [CHB]; Japanese from Tokyo, Japan [JPT]); and European (Utah residents with ancestry from Northern and Western Europe [CEU]) ancestry.

- **Genome-wide imputation grid** was selected using Affymetrix's imputation aware marker choice algorithms[4,5] to provide genome-wide coverage in Caucasian European populations of common (estimated minor allele frequency [EMAF] ≥5%) markers (using the EUR panel defined as the GBR, CEU, FIN, IBS, and TSI samples from the 1000 Genomes Project).[6]
- **Genome-wide coverage of non-European populations** markers were added from Axiom Biobank Genotyping Array for improved coverage of non-European populations, including ASW, YRI, CHD, CHB, and JPT.
- **Genome-wide booster panel** was included to improve imputation coverage for AFR and CEU populations. Coverage of common variants (minor allele frequency [MAF] >2% in CEU and >5% in AFR) was improved by selecting additional genome-wide variants.
- **Compatibility markers** were included covering ancestry informative markers; fingerprint, mitochondrial, and Y chromosome markers; and miRNA binding sites/target regions. These markers allow for sample tracking and optimization and standardization of genotyping quality control.

While the genomic coverage has been optimized for major HapMap populations, the advanced array design also provides comprehensive coverage of markers relating to transplantation research.

Module content from UK Biobank Axiom Array

- Genes in the **HLA** region of chromosome 6 **and KIR** region of chromosome 19 are known to be important in immune response, but are difficult to directly assay on commercial arrays. Markers have been added to the array to facilitate imputation of these alleles.
- **Phenotype associations content** was selected directly from, or as tags for, markers in the National Human Genome Research Institute (NHGRI) *Catalog of Published Genome-Wide Association Studies*, up to mid 2013.
- **Known copy number variants (CNVs)** were selected to cover CNV regions and evaluate genomic structural elements. Markers were added to ensure dense coverage within the CNV region, and assays were included for known breakpoints, where possible. Markers were also added to ensure coverage of a small number of CNVs, which have shown suggestive evidence of association with lung function, and a small number of candidate CNV regions for which there was interest to test association with lung function and related phenotypes. Additional CNV and copy number polymorphism (CNP) variants are included in the functional variant module.
- **Expression quantitative trait loci (eQTL)** were supplied for UK Biobank Axiom Array from the GEUVADIS project,[7] together with eQTL for several other discovery projects, including the MuTHER project, the ALSPAC project, and the GENCORD project. These markers were included to support mapping functional non-coding variations to identify genetic markers associated with gene transcription variability and differential gene expression.
- **Lung tissue/pulmonary function** markers with established or putative association with lung function, lung disease (including asthma, cystic fibrosis, chronic obstructive pulmonary disease, idiopathic pulmonary fibrosis, and lung cancer), smoking behavior, or any combination of these, were included on the array.

Expanded major histocompatibility complex (MHC) and KIR content was chosen in addition to the HLA module listed above in order to improve imputation of major HapMap populations, including African (ASW, YRI), Asian (CHB, CHD, JPT), and European (CEU) populations.

- **Multi-ethnic, HLA haplotype tagging single-nucleotide polymorphisms (SNPs)**[8] that are in linkage disequilibrium (LD) with HLA alleles were included for improved imputation of HLA alleles in multi-ethnic populations.
- A set of markers from the **Type 1 Diabetes Genetic Consortium (T1DGC) HLA imputation panel**[9] was chosen for direct tiling, tagging, or both by LD for HLA imputation.
- **Validated, non-redundant MHC SNPs** were selected from existing genotyping platforms, including Infinium® MetaboChip and ImmunoChip.[10]

Transplant-specific content module was customized for metabolic and pharmacogenomic markers that are known and potentially relevant in transplantation.

- **Pharmacogenomic** absorption, distribution, metabolism, and excretion (ADME) markers were selected from the Pharmacogenomics Knowledgebase[11] with known relevance to drug absorption, metabolism, excretion, and toxicity. Additional markers were selected based on the genotyping results for candidate genes and pathways identified by the Deterioration of Kidney Allograft Function (DeKAF) study. Coverage of markers involved in the metabolism of ISTs and other transplant-related therapeutics were also included in this module.
- **Candidate genes** curated from a PubMed search of over 600 transplant-related genetic association studies were included. These markers were selected with associations in solid organ transplantation outcomes, specifically: liver, lung, heart, and kidney. Covering over 90,000 polymorphic loci, these SNPs were selected to maximize coverage across CEU, YRI, and ASN populations. In addition to transplant outcome, transplant associations (e.g., new-onset diabetes after transplantation) and response to transplant prescription therapies were also evaluated.

Functional variants of putative coding and non-coding LoF variants were selected when represented by no less than five minor allele counts (MACs) in whole-exome sequencing reference datasets.

- **Exonic and coding** markers were selected from Axiom Biobank Genotyping Array, including putative exonic or coding single-nucleotide variants (SNVs).
- **LoF** markers were selected from Axiom Biobank Genotyping Array as well as human disease mutation and exome databases.
- **Untranslated region (UTR)** markers were chosen for potential effect of functional gene expression. Using an MAF of >1% for European populations and >5% for African populations, variants were selected to cover 5′ and 3′ UTRs as defined by either RefSeq or Ensembl.
- ***A priori* association** variants were selected based on calculated genome-wide significance of $P < 5 \times 10^{-8}$, as reported in NHGRI GWAS catalog.
- **CNVs and CNPs** were chosen that cover 2,200 curated CN regions. The median number of markers selected to cover each CNV region varied from 3–122, with a median gap size of 196–2,602 base pairs, depending on the size of the CNV.

Table 1: Categories of markers on Axiom® Transplant Genotyping Array.

Category/Markers of specific interest	Number of markers
Module content from UK Biobank Axiom® Array	
HLA and KIR	8,894
Phenotype associations	8,136
Known CNVs	2,369
eQTL	17,115
Lung tissue/pulmonary function	8,645
Expanded MHC and KIR content	
Multi-ethnic, HLA haplotype tagging SNPs	421
T1DGC HLA imputation panel	4,794
Validated, non-redundant MHC SNPs	13,732
Transplant-specific content	
Pharmacogenomic	9,500
Candidate genes	23,800
Functional variants	
Exonic/coding variants	168,000
LoF	28,128
UTR	184,000
A priori associations	8,136
CNVs/CNPs	27,370
GWAS markers	
Genome-wide imputation grid	296,000
Genome-wide coverage for non-European populations	50,000
Genome-wide booster	135,000
Compatibility markers	8,000
Total number of markers	782,000

Specifications

Genotyping performance has been evaluated on 270 samples, including samples from the International HapMap Project, against stringent quality control metrics covering average sample call rate, sample concordance, and reproducibility. See Table 2 for performance metrics and validation data.

Table 2: Performance metrics for Axiom® Transplant Genotyping Array.

Metric	Specification	HapMap
Number samples		270
Sample pass rate	≥95%	100%
Average call rate	≥99%	99.71%
Reproducibility	≥99.8%	99.96%
Average HapMap concordance	≥99.5%	99.77%

Analysis workflow for Axiom Transplant Genotyping Array

The advanced analysis workflow for Axiom Transplant Genotyping Array requires the use of the .r3 version of the analysis package. This package is available for download from within Axiom™ Analysis Suite or from the Technical Documentation tab of the Axiom Biobank Genotyping Arrays product page. This workflow is an advanced analysis technique that utilizes SNP-specific priors (SSPs) to provide the greatest flexibility in finding the most informative content from each study. *Axiom Genotyping Solution Data Analysis Guide* (P/N 702961) details the Best Practices Workflow; the *Analysis Note for Axiom Transplant Genotyping Array* (P/N 703407) details the additional analysis options available for this array.

References

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Ordering information

Part number	Description	Details
902865	Axiom® Transplant Genotyping Array	Contains one 96-array plate
000854	Axiom® Biobank Plus Genotyping Array	Contains one Axiom Biobank Genotyping Array plus up to 50,000 additional custom markers
901606	Axiom® GeneTitan® Consumables Kit	Includes all GeneTitan Instrument consumables required to process one Axiom array plate
901758	Axiom® 2.0 Reagent Kit	Includes all reagents (except isopropanol) required to process 96 DNA samples

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