PharmacoScan Solution

Preemptive genotyping of known pharmacogenomics markers in a single assay

Personalized medicine is becoming a reality with the advancement of technologies that allow us to understand the common variation in genes coding for drug metabolism and drug transporters. However, attrition of drug candidates in the pharmaceutical industry continues to make the drug discovery process costly and lengthy. This attrition is commonly due to poor pharmacokinetics and toxicity—two risks that can be strongly influenced by the variability in drug responses due to genetic variation. The process of dissecting environmental factors, physiological variables, and genetic characteristics (pharmacogenetics) is complicated by the large numbers of drug-metabolizing enzymes that contribute to the processing of most drugs.

A molecular assay designed for multiethnic populations that targets functional variations in all the key genes involved in the absorption, distribution, metabolism, and excretion (ADME) of commonly prescribed medications can help reduce the lengthy timelines and complexities within the drug discovery process. The Applied Biosystems™ PharmacoScan™ Solution offers such capability by providing broad coverage of industry-relevant, multiethnic content, and the capability to address markers in complex genes on a platform that has superior lot-to-lot reproducibility. The PharmacoScan Solution allows clinical researchers to gain valuable insight into an individual's ability to process drugs based upon high, moderate, low, and preliminary scientific evidence.

Key features

- 4,627 markers in 1,191 genes of known pharmacogenomic value
- Comprehensive content of known pharmacogenomic value including phase I and phase II enzymes, regulatory and modifier genes, drug target genes, and phase III transporter genes
- Genotyping of highly predictive markers in genes, including *GSTM1*, *CYP1A2*, *CYP2D6*, *CYP2B6*, *CYP2A6*, *SULT1A1*, *CYP2C19*, and *CYP2C9* that are in highly homologous regions
- Copy number variation (CNV) analysis for copy number states ranging from 0 to >3 for important ADME genes
- Star allele and translation tables for key actionable genes

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Content description

Content of the Applied Biosystems™ PharmacoScan™ array (Figure 1) includes:

- Core functional pharmacogenomic content, including markers from the Clinical Pharmacogenomics Implementation Consortium (CPIC) guidelines, the Pharmacogenomics Knowledge Base (PharmGKB) markers in Very Important Pharmacogenes (VIP), PharmGKB markers with clinical annotations, and PharmaADME core markers
- Nearly all markers from the Applied Biosystems™ DMET™ Plus Solution (1,936 genetic variants across 231 relevant genes) [1–5]
- Human leukocyte antigen (HLA) markers associated with drug reactions
- Markers for killer cell immunoglobulin-like receptors (KIR), human ancestry identification, and sample identification and tracking
- Pharmacogenetics and ADME markers in genes targeted for European populations drawn from the Applied Biosystems™ UK Biobank Axiom™ Array

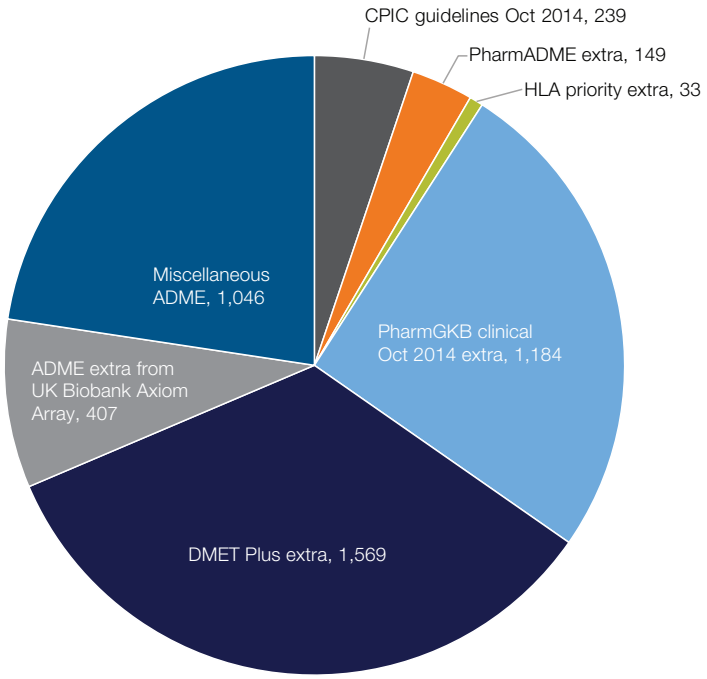

Figure 1. Distribution of consortia and sources used to select the ADME markers for the PharmacoScan Solution.

Continuity of content for pharmacogenomics trials

Drug development research and clinical trials are typically conducted over several years. These long-term efforts require a platform that can offer multiyear availability of 100% of the specific array content for the entire timeline of the initiative. Unlike bead-based technologies that experience batch-to-batch variability and single nucleotide polymorphism (SNP) dropouts with each manufacturing batch, the photolithography manufacturing technology used in the PharmacoScan array enables 100% fidelity and helps ensure that all markers are present on every manufacturing batch—addressing a major concern for such long-term efforts. The PharmacoScan Solution utilizes the same chemistry from the Axiom array and Applied Biosystems™ GeneTitan™ Multi-Channel (MC) Instrument, a platform used worldwide by genetic researchers for the efficient workflow, high throughput, and reproducibility of results critical to multiyear data collection and analysis efforts.

PharmacoScan assay

Pharmacogenomics research has often required the use of more than one molecular assay, increasing the cost and time to result. The PharmacoScan assay helps overcome this concern by interrogating SNPs, insertions or deletions (indels), and CNVs in a single-assay workflow. Key markers in pseudogenes like *CYP2D6* are amplified by multiplex PCR (mPCR) in the assay.

The PharmacoScan Solution includes the arrays, reagents, processing consumables, and analysis software needed for processing 88 samples on four 24-format array plates or 94 samples on each 96-format array plate. Each 24-format array plate can process 22 samples and 2 controls, and each 96-format array plate can process 94 samples and 2 controls.

Analysis software and results

Data generated using the PharmacoScan array is analyzed with Applied Biosystems™ Axiom™ Analysis Suite software. The output includes genotypes, as well as output of star allele and translation tables. The analysis uses the Applied Biosystems™ Axiom™ Best Practices Workflow, together with SNP-specific priors (SSPs), to provide the greatest flexibility for genotyping pharmacogenomic markers in each set of 22 or 94 samples. Copy number (CN) results are also presented in the software. This includes CN 0/1/2+ for *UGT2B17*, *GSTT1*, *GSTM1*, *CYP2A6* (3 regions), and 0/1/2/3+ for *CYP2D6* (3 regions).

The translation tables in the software translate the genotype calls of an important subset of SNPs to functional allele calls using standardized nomenclature wherever possible, enabling the following functions:

- Quick identification of possible rare alleles or missing data
- Identification of haplotype and SNP-level sequence variation in the test samples
- Annotation of reported genotypes across translated SNPs to indicate genomic, mRNA, or amino acid changes resulting from any observed variation
- Prediction of general gene activity based on detected diplotypes

The databases used to curate the allele translation gene tables include: (i) PharmGKB—Stanford University pharmacogenomics reference database, (ii) Karolinska *CYP450* gene standard nomenclature, (iii) Database of NAT genes (Democritus University of Thrace), (iv) Database of UGT genes, (v) Drug interaction database (University of Indiana), and (vi) PubMed—online National Library of Medicine publication database. The Axiom Genotyping Solution Data Analysis Guide (P/N 702961) details the Best Practices Workflow.

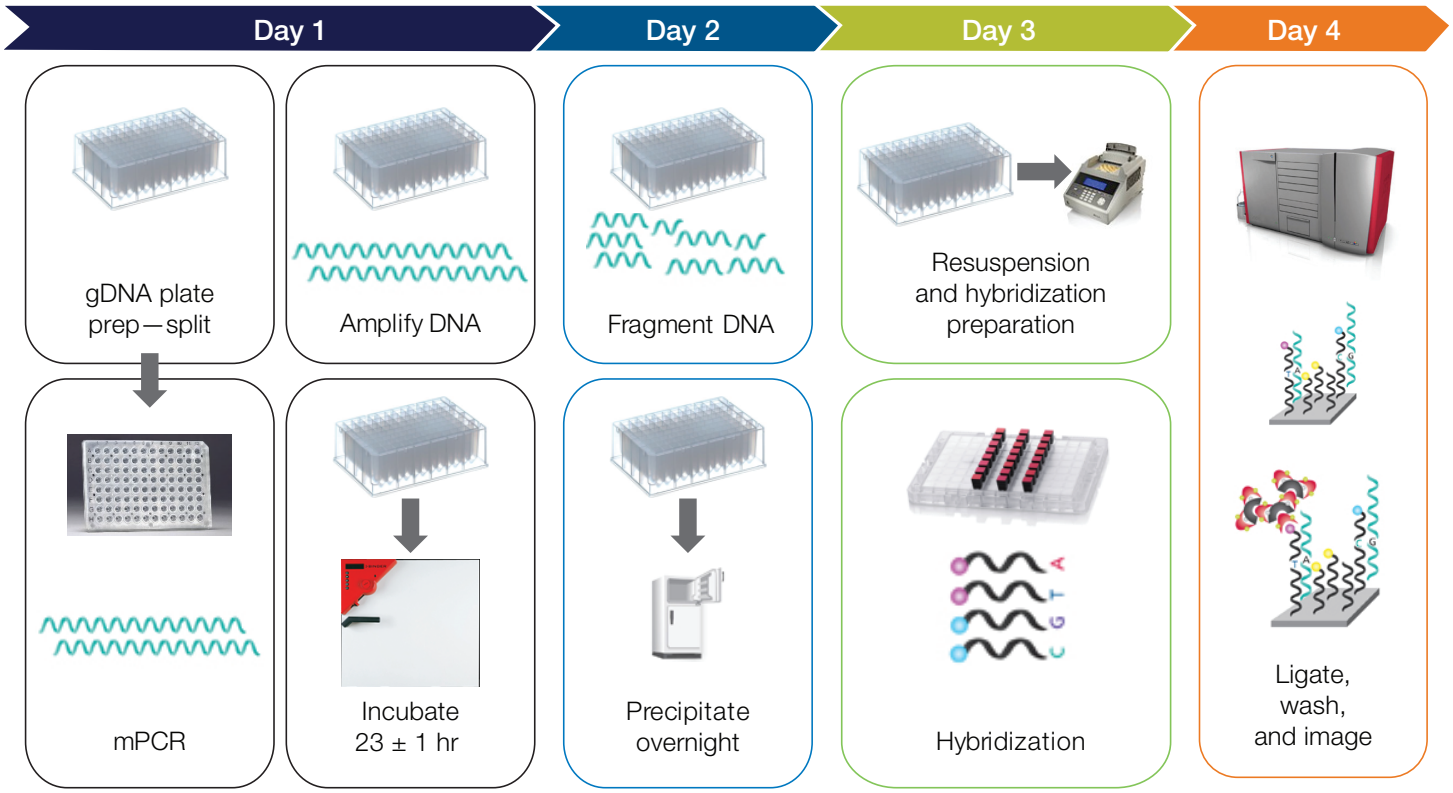

Figure 2. The PharmacoScan assay workflow.

The PharmacoScan Solution was evaluated at three independent sites as well as being validated at Thermo Fisher Scientific. The data were analyzed using the Axiom Best Practices Workflow. Results from each of the sites is presented in Table 1. Example of cluster plots and copy number results are shown in Figures 3 and 4, respectively.

Table 1. Data generated using the PharmacoScan assay at four independent sites.

Genotype attribute	Specification	Site 1	Site 2	Site 3	Thermo Fisher Scientific
Call rate across ADME genes	≥99.00%	99.95%	99.94%	99.92%	99.95%
Concordance across ADME genes	≥99.60%	99.85%	99.76%	99.80%	99.97%
Heterogeneous concordance across ADME genes	≥98.00%	99.60%	99.58%	99.41%	99.97%

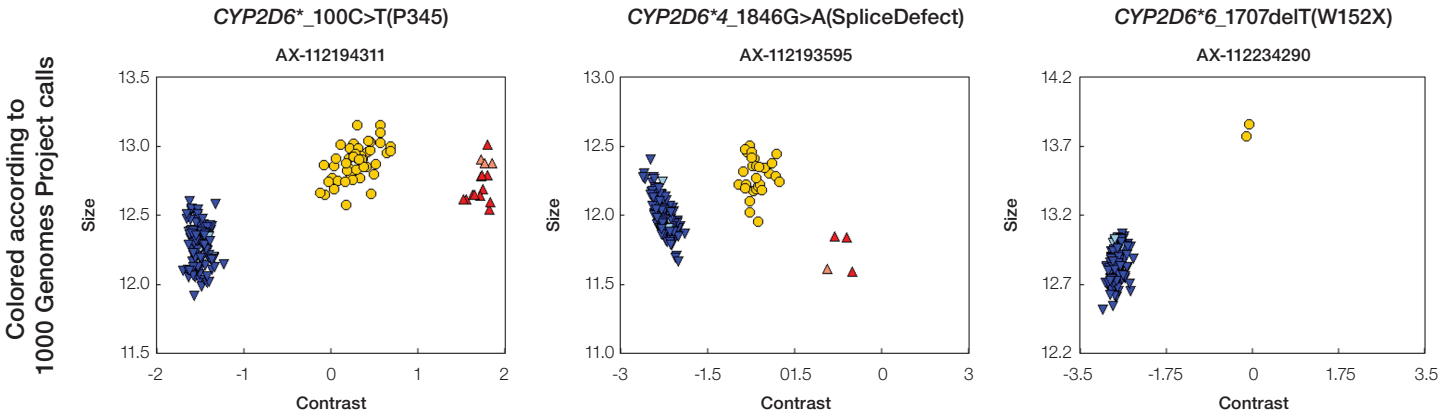

Figure 3. Plots represent cluster plots in contrast versus size space for *CYP2D6*10*, *CYP2D6*4*, and *CYP2D6*6* alleles. Calls are colored according to guidelines for 1000 Genomes Project representation.

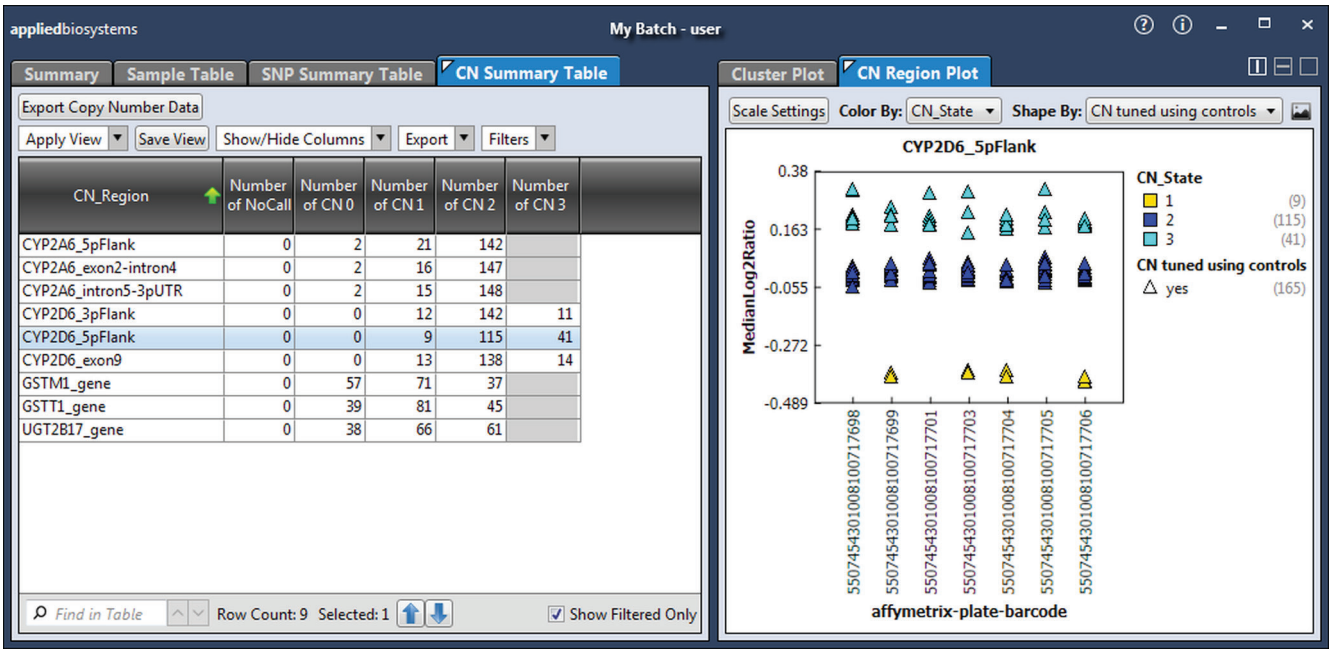

Figure 4. Capabilities of the PharmacoScan Solution for copy number analysis.

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References

1. Deeken J (2009) The Affymetrix DMET platform and pharmacogenetics in drug development. *Current Opinion in Molecular Therapeutics* 11(3):260–268.
2. Tremaine L, et al. (2015) The role of ADME pharmacogenomics in early clinical trials: perspective of the Industry Pharmacogenomics Working Group (I-PWG). *Pharmacogenomics* 16(18):2055–2067.
3. Wakil SM, Nguyen C, Muiya NP, et al. (2015) The Affymetrix DMET Plus platform reveals unique distribution of ADME-related variants in Ethnic Arabs. *Disease Markers* 2015:542543.
4. Medhasi S, Pasomsub E, Vanwong N, et al. (2016) Clinically relevant genetic variants of drug-metabolizing enzyme and transporter genes detected in Thai children and adolescents with autism spectrum disorder. *Neuropsychiatric Disease and Treatment* 12:843–851.
5. Jackson JN, et al. (2016) A comparison of DMET Plus microarray and genome-wide technologies by assessing population substructure. *Pharmacogenetics and Genomics* 26(4):147–153.

Ordering information

Product	Description	Quantity	Cat. No.
PharmacoScan Assay Kit	Includes four 24-array plates, PharmacoScan reagents, controls, and GeneTitan Multi-Channel Instrument consumables for running 22 samples on each plate	88 samples	903010TS
	Includes one 96-array plate, PharmacoScan reagents, controls, and GeneTitan Multi-Channel Instrument consumables for running 94 samples	94 samples	903026
PharmacoScan Training Kit	Includes four 24-array plates and two DNA plates for training, PharmacoScan reagents, controls, and GeneTitan Multi-Channel Instrument consumables for running 22 samples on each plate	88 samples	903011TS
	Includes two 96-array plates and two DNA plates for training, PharmacoScan reagents, controls, and GeneTitan Multi-Channel Instrument consumables for running 94 samples on each plate	188 samples	913027
PharmacoScan Reagent Kit	Includes PharmacoScan reagents and controls required to run four 24-array plates with 22 samples on each plate	88 samples	902908TS
	Includes PharmacoScan reagents and controls required to run one 96-array plate with 94 samples	94 samples	913025

Find out more at **thermofisher.com/microarrays**

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