The GeneChip® Gene 1.0 ST Array System offers a more complete and accurate view of total transcription activity at each genomic locus than classical 3’-based microarrays. Using a subset of Exon 1.0 ST Array probes that focus on well-annotated content, Gene 1.0 ST Arrays provide a cost-effective solution for whole-transcript (WT) coverage. This affordable format makes Gene 1.0 ST Arrays ideally suited for new microarray users, as well as researchers interested in cost-effective “gene-level” analysis based on whole-transcript coverage. To distinguish between alternatively spliced transcripts, the Affymetrix whole-transcript portfolio includes Exon 1.0 ST Arrays, which can also be used to perform gene-level analysis.

Gene 1.0 ST Arrays contain the most up-to-date content of well-annotated genes, and data is analyzed using a simple workflow. Gene 1.0 ST Arrays are part of a complete solution for gene expression analysis that includes reagents, instrumentation and basic analysis software.

Introduction

The GeneChip® Gene 1.0 ST Array System offers the most advanced and cost-effective gene expression profiling option for whole-transcript coverage. Probes are distributed across the full length of the gene, providing a more complete and accurate picture of overall gene expression.

Most 3’-based expression array designs have probes localized to the extreme 3’ end of the gene, and classical 3’ assays typically rely on priming from a transcript’s poly-A tail. Therefore, some genes may not be accurately represented on a classical 3’-based expression array in the following situations:

- Alternative splicing at the 3’ end of the gene
- Utilization of a novel 3’ terminal exon
- Alternative polyadenylation sites
- Nonpolyadenylated messages
- Truncated transcript
- Degraded samples
- Genomic deletions

These transcripts have been shown to play important biological roles and can now be detected as part of a gene-level expression estimate using the Whole Transcript (WT) Assay approach of the Gene 1.0 ST Array System.

Additional features of the Gene 1.0 ST Array System include:

- A cost-effective expression profiling solution uniquely suited for new microarray users interested in whole-transcript coverage.
- Simplified informatics and analysis; gene-level analysis of multiple probes on different exons summarized into a single expression value that represents all transcripts from the same gene.
- A complete solution for gene expression analysis that includes WT Sense Target Labeling and Control Reagents, fluidics and scanning instrumentation and basic analysis software. The array contains the most up-to-date content of well-annotated genes, and data is analyzed using a simplified workflow.
- The WT Assay has been optimized for compatibility with the Exon 1.0 ST and Gene 1.0 ST Array Systems. The result is a single source for highly reproducible, robust and consistent reagents.

Complete reagent kits have been developed, validated and qualified specifically for Gene 1.0 ST Arrays. These reagent kits provide convenience as well as high-quality results and consistent data.

Using a random priming strategy, in combination with in vitro transcription-based linear amplification and a novel end-point fragmentation and labeling assay scheme, the WT Assay provides a robust method for target labeling.

The 100 ng total RNA labeling protocol is recommended for use with Gene 1.0 ST Arrays, supporting a wide range of applications with limited starting material (no RiboMinus required).
Comprehensive Coverage of the Entire Gene Locus

The Gene 1.0 ST Array System uses a perfect match (PM)-only design with probes that hybridize to sense targets. Background is estimated using a set of approximately 17,000 generic background probes. Standard poly-A controls and hybridization controls are represented on the arrays to allow convenient troubleshooting along the entire experimental process.

Gene 1.0 ST Arrays use a subset of probes from Exon 1.0 ST Arrays and cover only well-annotated content (see Figure 2). Like Exon 1.0 ST Arrays, gene-level analysis of multiple probes on different exons is summarized into an expression value representing all transcripts from the same gene.

The GeneChip® Human Gene 1.0 ST Array, the first array launched in the Gene Array family, interrogates 28,869 well-annotated genes with 764,885 distinct probes. The design of the Human Gene 1.0 ST Array was based on the March 2006 human genome sequence assembly (UCSC Hg18, NCBI build 36) with comprehensive coverage of RefSeq, Ensembl and putative complete CDS GenBank transcripts. The Human Gene 1.0 ST Array has greater than 99 percent coverage of NM sequences present in the November 3, 2006, RefSeq database.

The GeneChip® Mouse and Rat Gene 1.0 ST Arrays are the latest additions to the Gene 1.0 ST Array family. The Mouse Gene 1.0 ST Array interrogates 28,853 well-annotated genes with 770,317 distinct probes. The design of the array was based on the February 2006 mouse genome sequence (UCSC mm8, NCBI build 36) with comprehensive coverage of RefSeq, putative complete CDS GenBank transcripts, all Ensembl transcript classes and syntenically mapped full-length mRNAs and RefSeq NMs from human and rat. The Mouse Gene 1.0 ST Array has 100 percent coverage of NM sequences present in the April 3, 2007, RefSeq database.

The GeneChip® Rat Gene 1.0 ST Array interrogates 27,342 well-annotated genes with 722,254 distinct probes. The design of the array was based on the November 2004 rat genome sequence (UCSC rn4, Baylor HGSC build 3.4) with comprehensive coverage of RefSeq, putative complete CDS GenBank transcripts, all Ensembl transcript classes and syntenically mapped full-length mRNAs and RefSeq NMs from human and mouse. The Rat Gene 1.0 ST Array has 99.98 percent coverage of NM sequences present in the April 3, 2007, RefSeq database (two NMs are LINE1 repeats that were deliberately excluded).

Gene 1.0 ST Array Performance

Comparable performance and strong concordance of the Human Gene 1.0 ST Array was observed against both the classical Human Genome U133 Plus 2.0 and the Human Exon 1.0 ST Arrays (see the Affymetrix White Paper, Human Gene 1.0 ST Array Performance, available at www.affymetrix.com/support). A Latin
Square spike-in experiment (see Figure 3) demonstrated comparable performance with the same high sensitivity and reproducibility for all three array design types. A heart-versus-brain tissue mixture study showed similar performance and strong concordance between the three array types.

The Mouse and Rat Gene 1.0 ST Arrays were designed using the same strategy as the Human Gene 1.0 ST Array, leveraging the performance and content optimizations and improvements made for the Human Gene 1.0 ST Array. Strong concordance of the Human, Mouse and Rat Gene 1.0 ST Arrays with their respective Exon 1.0 ST Arrays was observed in log ratio values for brain versus heart (see Figure 4).

Complete Instrumentation and Analysis Solution

For convenience and complete support, Gene 1.0 ST Arrays are provided as part of a comprehensive solution that includes Affymetrix instrumentation and analysis solutions:

- GeneChip® Fluidics Station 450 for complete walk-away array processing to obtain the highest level of reproducibility
- GeneChip® Scanner 3000 7G with the optional Autoloader for array image acquisition
- GeneChip® Hybridization Oven 640

Affymetrix offers several tools to help users with data analysis, including Expression Console™ software, the NetAffx™ Analysis Center and the Integrated Genome Browser (IGB). Expression Console software is an easy-to-use application that enables probe set summarization as well as preliminary data quality evaluation. A simple workflow enables the user to quickly analyze the data (see Figure 5). The resulting data can be further analyzed using software applications from Affymetrix GeneChip®-compatible™ software providers. A complete listing of these applications can be found at www.affymetrix.com.

The NetAffx Analysis Center, with regularly updated biological and functional annotations of probe sets, is the most comprehensive resource for array annotations and probe sequence information. Flexible query tools and external links allow researchers to...
**Figure 4:** Brain and heart samples were processed with RiboMinus and hybridized to Exon 1.0 ST and Gene 1.0 ST Arrays in triplicate. Brain and heart CEL files were normalized as a single group and RMA signals were obtained. The difference between the mean signal of brain and mean signal of heart samples is plotted below (exon array on x-axis, gene array on y-axis). Strong concordance was observed for human (A), mouse (B) and rat (C). The Pearson correlation coefficients were 0.956 for human (based on 16,482 probe sets), 0.974 for mouse (based on 16,233 probe sets) and 0.980 for rat (based on 8,161 probe sets).

**Figure 5:** The Gene 1.0 ST Array data analysis workflow is similar to the workflow for the 3’ gene expression analysis, utilizing Expression Console, third-party GeneChip-compatible software and annotation tools in the NetAffx Analysis Center and IGB.
explore genes and annotations of interest with significant breadth and depth. This resource makes it easy for researchers to interpret their microarray results and quickly design their downstream studies.

Researchers can also use IGB to visualize their results in a genomic context. Genomic annotations, including RefSeq sequences, SNPs and genomic locations from various sources, can be viewed alongside microarray gene expression signal data.

**Additional Support**

Available online at [www.affymetrix.com](http://www.affymetrix.com)

**WT Assay (for Human, Mouse and Rat)**


**White paper, Whole Transcript (WT) Sense Target Labeling Assay Performance**

**Tissue Mixture Array Performance Data Set, GeneChip Human Gene 1.0 ST Array**

**Tissue Panel Data Set for Human, Mouse and Rat Gene 1.0 ST Arrays**

**White Paper, Quality Assessment of Exon and Gene Arrays**

**White Paper, Human Gene 1.0 ST Array Performance**

**Technical Note, GeneChip® Human Gene 1.0 ST Array Design**

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**Table 1: The Gene 1.0 ST Array System.**

<table>
<thead>
<tr>
<th>Arrays</th>
<th>GeneChip® Human, Mouse and Rat Gene 1.0 ST Arrays</th>
</tr>
</thead>
<tbody>
<tr>
<td>Target Labeling Assay</td>
<td>GeneChip® Whole Transcript (WT) Sense Target Labeling Assay and Reagents</td>
</tr>
<tr>
<td>Fluidics Station</td>
<td>GeneChip® Fluidics Station 450</td>
</tr>
<tr>
<td>Scanner</td>
<td>GeneChip® Scanner 3000 7G or higher</td>
</tr>
<tr>
<td>Image Acquisition</td>
<td>GeneChip® Operating System (GCOS), Affymetrix GeneChip® Command Console (AGCC)</td>
</tr>
<tr>
<td>Quality Control and Gene-level Signal</td>
<td>Affymetrix Expression Console™ Software</td>
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**Specifications**

<table>
<thead>
<tr>
<th>Human</th>
<th>Mouse</th>
<th>Rat</th>
</tr>
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<tbody>
<tr>
<td>Number of arrays</td>
<td>1</td>
<td>1</td>
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<tr>
<td>Array format</td>
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<td>169</td>
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<tr>
<td>Feature size</td>
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<td>5 µm</td>
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<tr>
<td>Oligonucleotide probe length</td>
<td>25-mer probes</td>
<td>25-mer probes</td>
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<tr>
<td>Total number of distinct probes</td>
<td>764,885</td>
<td>770,317</td>
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<td>Interrogated strand</td>
<td>Sense</td>
<td>Sense</td>
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<tr>
<td>Resolution (number of probes per gene)</td>
<td>26 (median)</td>
<td>27 (median)</td>
</tr>
<tr>
<td>Estimated number of genes</td>
<td>28,869</td>
<td>28,853</td>
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<tr>
<td>Gene-level probe sets with Ensembl support</td>
<td>28,132</td>
<td>27,543</td>
</tr>
<tr>
<td>Gene-level probe sets with putative full-length transcript support (GenBank and RefSeq)</td>
<td>19,734</td>
<td>19,434</td>
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<td>Genome assembly</td>
<td>March 2006 (UCSC Hg 18; NCBI build 36)</td>
<td>February 2006 (UCSC mm8; NCBI build 36)</td>
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<tr>
<td>Positive controls (constitutively expressed genes)</td>
<td>1,195 putative exon-level probe sets from putative constitutive genes</td>
<td>1,324 putative exon-level probe sets from putative constitutive genes</td>
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<td>Negative controls</td>
<td>2,904 putative intron-level probe sets from putative constitutive genes</td>
<td>5,222 putative intron-level probe sets from putative constitutive genes</td>
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<td>Hybridization controls</td>
<td>bioB, bioC, bioD, cre</td>
<td>bioB, bioC, bioD, cre</td>
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<td>Background probes</td>
<td>Antigenomic Set</td>
<td>Antigenomic Set</td>
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<tr>
<td>Poly-A controls</td>
<td>dap, lys, phe, thr</td>
<td>dap, lys, phe, thr</td>
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<tr>
<td>Recommended amount of starting material</td>
<td>100 ng (without RiboMinus)</td>
<td>100 ng (without RiboMinus)</td>
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<td>Acceptable range of input amount</td>
<td>• 1 µg protocol: 1-2 µg (with Ribominus)</td>
<td>• 1 µg protocol: 1-2 µg (with Ribominus)</td>
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<tr>
<td></td>
<td>• 100 ng protocol: 100-300 ng (no Ribominus)</td>
<td>• 100 ng protocol: 100-300 ng (no Ribominus)</td>
</tr>
</tbody>
</table>
Ordering Information

GeneChip® Gene 1.0 ST Array System

**900652 GeneChip® WT Sense Target Labeling and Control Reagents, 30 reactions**
Contains:
- GeneChip® WT cDNA Synthesis and Amplification Kit
- GeneChip® WT Terminal Labeling Kit
- GeneChip® Sample Cleanup Module
- GeneChip® IVT cRNA Cleanup Kit
- GeneChip® Eukaryotic Poly-A RNA Control Kit, ~100 reactions
- GeneChip® Hybridization Control Kit

**901168 GeneChip® Mouse Gene 1.0 ST Array**
- Contains 2 Arrays

**901169 GeneChip® Mouse Gene 1.0 ST Array**
- Contains 6 Arrays

**901171 GeneChip® Mouse Gene 1.0 ST Array**
- Contains 30 Arrays

**901172 GeneChip® Rat Gene 1.0 ST Array**
- Contains 2 Arrays

**901173 GeneChip® Rat Gene 1.0 ST Array**
- Contains 6 Arrays

**901175 GeneChip® Rat Gene 1.0 ST Array**
- Contains 30 Arrays

**Array and Reagent Combination Kits, Human Gene 1.0 ST Array**

**901147**
- Contains 10 arrays and reagents for 10 reactions

**901146**
- Contains 30 arrays and reagents for 30 reactions

**Array and Reagent Combination Kits, Mouse Gene 1.0 ST Array**

**901178**
- Contains 10 arrays and reagents for 10 reactions

**901179**
- Contains 30 arrays and reagents for 30 reactions

**Array and Reagent Combination Kits, Rat Gene 1.0 ST Array**

**901176**
- Contains 10 arrays and reagents for 10 reactions

**901177**
- Contains 30 arrays and reagents for 30 reactions

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