

GeneChip® WT
Amplified Double-Stranded
cDNA Synthesis Kit



INTRODUCTION

The GeneChip® WT (Whole Transcript) Amplified Double-Stranded cDNA Synthesis Kit is optimized for use in the first steps of the GeneChip WT Amplified Double-Stranded Target Assay. This assay is designed to generate targets compatible with GeneChip® Tiling Arrays. When used in combination with GeneChip Tiling Arrays, the WT Amplified Double-Stranded Target Assay can provide unbiased transcript mapping results on a genome-wide scale.

Key characteristics of the WT Amplified Double-Stranded Target Assay:

- Robust assay and reagents for generating double-stranded cDNA targets, which are subsequently hybridized to GeneChip Tiling Arrays.
- Complementary reagent kits associated with the WT Amplified Double-Stranded Target Assay are available, including the GeneChip WT Double-Stranded DNA Terminal Labeling Kit (P/N 900812).
- An alternative assay may be used when target amplification is neither desired nor required. The unamplified version of the assay is supported by the GeneChip WT Double-Stranded cDNA Synthesis Kit (P/N 900813).

The WT Amplified Double-Stranded Target Assay schematic is shown in Figure 1. The RNA starting material is primed with oligonucleotides composed of random hexamers conjugated to a T7 RNA Polymerase promoter sequence. Following first- and second-strand cDNA synthesis, cRNA is generated by *in vitro* transcription. After a clean-up step, the amplified cRNA is split into three tubes, and an additional round of random-primed cDNA synthesis is performed. The resulting double-stranded DNA is ready to be fragmented and labeled with the GeneChip WT Double-Stranded DNA Terminal Labeling Kit. After labeling, the DNA fragments are hybridized to a GeneChip Tiling Array, washed, and stained in preparation for subsequent scanning and data analysis.

Detailed procedures for this assay are described in the *GeneChip® WT Double-Stranded Target Assay Technical Manual* (P/N 702179), available online at www.affymetrix.com.

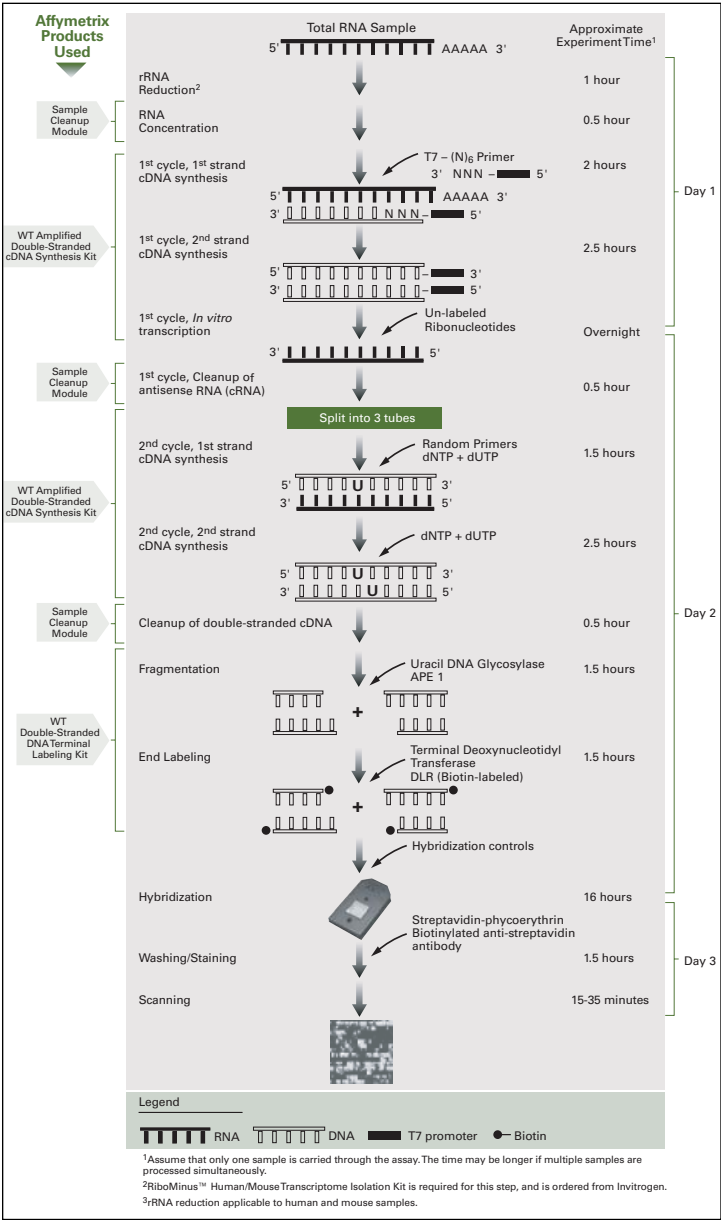


Figure 1. GeneChip® WT Amplified Double-Stranded Target Assay

ORDERING INFORMATION

P/N	Size	Kits
900811	10 Rxns	GeneChip® WT Amplified Double-Stranded cDNA Synthesis Kit

KIT CONTENTS

GeneChip® WT Double-Stranded cDNA Synthesis Module		
T7-(N) ₆ Primers, 2.5 µg/µL		20 µL
5X 1st Strand Buffer		140 µL
DTT, 0.1 M		70 µL
dNTP, 10 mM		9 µL
RNase Inhibitor		15 µL
SuperScript™ II		153 µL
MgCl ₂ , 1M		40 µL
DNA Polymerase I		42 µL
RNase H		17 µL
Random Primers, 3 µg/µL		30 µL
dNTP+dUTP, 10 mM		68 µL
RNase-free Water		4,440 µL

GeneChip® WT cDNA Amplification Kit		
10X IVT Buffer		50 µL
IVT NTP Mix		200 µL
IVT Enzyme Mix		50 µL
IVT Control		10 µL

ADDITIONAL AFFYMETRIX® REAGENTS ORDERED SEPARATELY

Kit	P/N	Size
GeneChip® WT Double-Stranded DNA Terminal Labeling Kit	900812	30 Reactions
GeneChip® Sample Cleanup Module	900371	30 Reactions
GeneChip® Control Oligo B2, 3 nM	900301	30 Reactions
GeneChip® WT Double-Stranded cDNA Synthesis Kit	900813	30 Reactions

STORAGE

Kit should be kept at -20°C in a non-frost-free freezer. Performance of the kit components has been shown to be unaffected for up to 8 freeze-thaw cycles.

FUNCTIONAL TESTING

Each lot of reagents is functionally tested using the GeneChip assay following the recommended protocol.

SAFETY INFORMATION


Material Safety Data Sheet(s) is sent only on the first shipment, unless a substantial revision occurs. If the product is a kit or is supplied with more than one material, please refer to the MSDS for each component for hazard information.

PATENTS

Use of this kit in accordance with these instructions in preparing targets for GeneChip® probe arrays may be covered by U.S. patent numbers 5,545,522; 5,716,785; 5,891,636 and 6,291,170. Those who do not use this kit to prepare targets for GeneChip probe arrays may be required to obtain a license under these patents.

Certain components of this kit or combinations thereof may be covered by U.S. patent numbers 5,256,555; 6,586,218 and 6,586,219, assigned to Ambion Inc.

TRADEMARKS

Affymetrix®, , GeneChip®, HuSNP®, GenFlex®, Flying Objective™, CustomExpress®, CustomSeq®, NetAffx™, 'The Way Ahead™', 'Tools to Take You As Far As Your Vision®', 'Powered by Affymetrix™', and 'GeneChip-compatible™' are trademarks of Affymetrix, Inc.

SuperScript™ is a trademark of Invitrogen Corporation.

CONTACT INFORMATION

Affymetrix, Inc.

3380 Central Expressway
Santa Clara, CA 95051 USA
E-mail: support@affymetrix.com
Tel: 1-888-362-2447 (1-888-DNA-CHIP)
Fax: 1-408-731-5441

Affymetrix UK Ltd

Voyager, Mercury Park,
Wycombe Lane, Wooburn Green,
High Wycombe HP10 0HH
United Kingdom
E-mail: supporteurope@affymetrix.com
UK and Others Tel: +44 (0) 1628 552550
France Tel: 0800919505
Germany Tel: 01803001334
Fax: +44 (0) 1628 552585

Affymetrix Japan, K. K.

Mita NN Bldg, 16 Floor
4-1-23 Shiba, Minato-ku
Tokyo 108-0014, Japan
supportjapan@affymetrix.com
Tel: +81-(0)3-5730-8200
Fax: +81-(0)3-5730-8201

Please visit our web site for international distributor contact information.

www.affymetrix.com

LIMITED LICENSE

The user of this kit and its components (the "Kit") is granted a limited license to the abovementioned patents and all u.s. and foreign patents and/or patent applications claiming priority from the patents, as follows:

User restrictions for research field:

This limited license permits the person or legal entity to which this kit has been provided to use the kit only with a nucleic acid array manufactured and sold by Affymetrix Inc. (An "Array"), and to use the data generated by use of the kit, only for its internal research purposes to identify and study the presence and/or levels of expressed nucleic acid sequences, and/or polymorphic variations in nucleic acid sequences, in the ordinary course of buyer's normal internal research activities, including clinical trials. In addition, buyer agrees and understands it is not licensed to (a) provide data derived from the use of the kit to or on behalf of any third party, whether on an exclusive or a non-exclusive basis, for consideration; or (b) use the kit or data therefrom in a clinical diagnostic setting where the data from an individual's sample is used to make a decision on behalf of an individual and the result is reported to such individual or their caregiver. neither affymetrix nor its licensors grants any other licenses, express or implied, to permit the manufacture, use, sale or importation of the kit, any array, any nucleic acids on the array, or any data generated from use of the array or the kit.

User restrictions for services field:

This limited license permits the person or legal entity to which this kit has been provided to use the kit only with a nucleic acid array manufactured and sold by Affymetrix Inc. (an "Array"), and to use the data generated by use of the kit, only for its internal research purposes to identify and study the presence and/or levels of expressed nucleic acid sequences, and/or polymorphic variations in nucleic acid sequences, in the ordinary course of buyer's normal internal research activities, including clinical trials, and to sell the data so generated for the exclusive use of a customer of the buyer in the ordinary course of such customer's internal research activities, including clinical trials. In addition, buyer agrees and understands, and will obtain the agreement of buyer's customer, that neither buyer nor any customer of buyer is licensed to use the kit or data therefrom in a clinical diagnostic setting where the data from an individual's sample is used to make a decision on behalf of an individual and the result is reported to such individual or their caregiver. Except as specifically described in this label license, neither affymetrix nor its licensors grants any other licenses, express or implied, to permit the manufacture, use, sale or importation of the kit, any array, any nucleic acids on the array, or any data generated from use of the kit.

User restrictions for commercial databases field:

This limited license permits the person or legal entity to which this kit has been provided to use the kit only with a nucleic acid array manufactured and sold by Affymetrix Inc. (an "array"), and to use the data generated by use of the kit, only for its internal research purposes to identify and study the presence and/or levels of expressed nucleic acid sequences, and/or polymorphic variations in nucleic acid sequences, in the ordinary course of buyer's normal internal research activities, including clinical trials, and to sell the data so generated for the nonexclusive use of customers of the buyer in the ordinary course of such customers' internal research activities, including clinical trials. In addition, buyer agrees and understands, and will obtain the agreement of buyer's customer, that neither buyer nor any customer of buyer is licensed to use the kit or data therefrom in a clinical diagnostic setting where the data from an individual's sample is used to make a decision on behalf of an individual and the result is reported to such individual or their caregiver. except as specifically described in this label license, neither Affymetrix nor its licensors grants any other licenses, express or implied, to permit the manufacture, use, sale or importation of the kit, any array, any nucleic acids on the array, or any data generated from use of the kit.

COPYRIGHT

©2005 Affymetrix, Inc. All rights reserved.