

High Quality RNA for Use with Microarrays

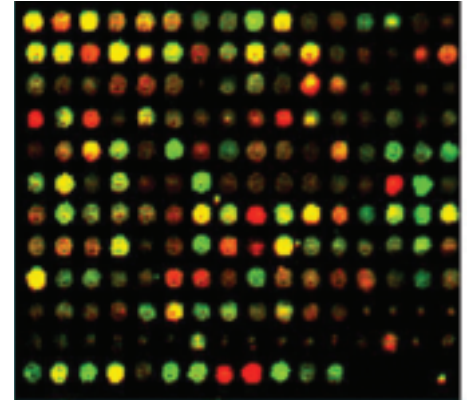
A recent journal article¹ coauthored by scientists from the University of Washington, Affymetrix, and the Institute for Systems Biology, which is a nonprofit research institute founded by Dr. Leroy Hood, describes the use of an *E. coli*, whole-genome microarray to more completely identify all *E. coli* RNAs transcribed under different growth conditions. EPICENTRE's MasterPure™ Complete DNA and RNA Purification Kit was the method of choice for isolating total RNA for reverse transcription and cDNA synthesis.

A current challenge for genome scientists is the gap between the number of sequenced genomes and the number of completely annotated and published genomes. A major reason for the gap is the difficulty of annotating (or interpreting) raw sequence data. The bioinformatics algorithms that convert primary sequence data to predicted genes do not address un-

translated regions (UTRs), including small RNAs that may have regulatory functions.

To avoid those limitations, these researchers developed a new approach to identify transcribed, intergenic regions in prokaryotes. High density oligonucleotide probe arrays (Affymetrix) covering the whole genome of *E. coli*, were hybridized with either fragmented, labeled genomic DNA or cDNA, which was prepared from RNA purified with the MasterPure Kit. The arrays assay both sense and antisense strands from intergenic regions, and sense strands from coding regions. Using this technique, they detected 4,052 coding transcripts and identified 1,102 additional transcripts from intergenic regions and 317 new transcripts of unknown function.

Thus, EPICENTRE's MasterPure™ Complete DNA and RNA Purification Kit is an effective tool in microarray technology.



Reference

1. B. Tjaden et al. (2002) *Nucl. Acids Res.* **30**, 3732.

For more information on MasterPure™ DNA and RNA Purification Kits, please see the center insert.

MasterPure™ Genomic DNA for *Bacillus anthracis* Analysis

A number of laboratories are working to develop quick, reliable, and sensitive assays for *Bacillus anthracis*, the causative agent of anthrax. Polymerase Chain Reaction (PCR) provides a good basis for rapid and specific analysis of genomic characteristics and is being utilized to detect and identify *Bacillus* species and strains. Performing PCR on a large number of clinical samples requires a fast, consistent method of genomic DNA purification. Here are two examples of laboratories using EPICENTRE's MasterPure™ DNA Purification Kit to prepare *Bacillus* DNA for their assays.

The international Laboratory Response Network (LRN) was established to prepare for, and respond to, potential bioterrorism threats. One of the first projects for this group was to develop rapid and highly specific assays to identify bioterrorism agents, including *B. anthracis*.¹ The LRN PCR assays for *B. anthracis* use real-time PCR and primer/probe sets against 3 genomic targets: pXO1, the 182-kb virulence plasmid; pXO2, the 96-kb virulence

plasmid; and a region on the chromosome. For assay development, reaction template came from vegetative cells or from spores. The lab uses EPICENTRE's MasterPure DNA Purification Kit to purify the DNA from vegetative cells, after a lysozyme/lysozyme pre-treatment. Spores were used directly in the assays.

In another approach to *Bacillus* identification, Radnedge et al.² used amplified fragment length polymorphism (AFLP) analysis, which provides a rapid method to measure phylogenetic distances. They effectively applied suppression subtractive hybridization (SSH) to develop and validate an AFLP analysis capable of generating highly specific DNA fingerprints of *Bacillus* species and strains. The lab purifies *Bacillus* genomic DNA from liquid, overnight cultures with EPICENTRE's MasterPure DNA Purification Kit.

References

1. Hoffmaster, A.R. et al. (2002) *Letters in Emerg. Infect. Dis.* **8**, 1178.
2. Radnedge, L. et al. (2003) *Appl. Environ. Microbiol.* **69**, 2755.

www.epicentre.com/purification.asp

MasterPure™ Complete DNA & RNA Purification Kit (for isolating TNA, DNA, or RNA)

MC89010 10 Purifications
(10 TNA or 10 DNA or 5 RNA)
MC85200 200 Purifications
(200 TNA or 200 DNA or 100 RNA)

Contents:

Red Cell Lysis Solution, Tissue and Cell Lysis Solution, MPC Protein Precipitation Reagent, 2X T&C Lysis Solution, TE Buffer**, RNase A, RNase-Free DNase I, Proteinase K, and 1X DNase Buffer.

**Not provided with the 10 purification kit (cat. no. MC89010).

MasterPure™ DNA Purification Kit (for isolating TNA or DNA)

MCD85201 200 Purifications

Contents:

Red Cell Lysis Solution, Tissue and Cell Lysis Solution, MPC Protein Precipitation Reagent, 2X T&C Lysis Solution, TE Buffer, RNase A, Proteinase K

MasterPure™ RNA Purification Kit (for isolating RNA only)

MCR85102 100 Purifications

Contents:

Red Cell Lysis Solution, Tissue and Cell Lysis Solution, MPC Protein Precipitation Reagent, 2X T&C Lysis Solution, TE Buffer, RNase-Free DNase I, Proteinase K, 1X DNase Buffer