

# High-throughput Genotyping Assay

## Affymetrix Uses Liquidator® 96



**Affymetrix's Research Services Laboratory (ARSL) in Santa Clara, California, provides highthroughput genotyping services to customers performing large-scale microarray-based studies. ARSL's goal is to provide quality data and minimize the time needed to complete genotyping studies. Technicians process samples in 96-well plates using the Liquidator 96 for efficient high-throughput studies.**

Since it was introduced in 1985, PCR has revolutionized life science labs with applications such as qPCR, genotyping assays and whole genome sequencing, adding new dimension to our understanding of gene function and structure.

PCR enables researchers to amplify very small amounts of starting template, but strict adherence to assay set up regimes is necessary due to the increased risk of errors. For optimal results, each of the liquid handling steps involved in adding sample and reagents to the 96-well plates must be tightly managed. The results of a genomic assay depend on many factors, including:

- Cleanliness of the lab environment, tools and accessories
- Purity of starting sample and reagents
- Choice of instrumentation and liquid handling tools
- Researcher's expertise in generating reproducible data

Many life science labs have adapted PCR applications to fit their specific research needs. For example, genotyping assays using microarray technologies have helped identify many disease specific genes. The ARSL offers a whole-genome genotyping assay that can provide insight into an organism's genetic make-up and gene expression profile. Preparing high-quality DNA for genotyping studies using PCR is an important step in the assay. The following protocol outlines ARSL's PCR plate set up method for processing samples on a whole-genome genotyping assay.

### Aliquot ligated DNA to the PCR plates using the Liquidator 96

1. Dilute the ligated DNA sample by adding reagent to a 96-well plate using the Liquidator 96.
2. Transfer 10  $\mu$ l each of diluted ligated sample from each row to the corresponding well of each PCR reaction plate.  
For example (Figure 1): transfer 10  $\mu$ L of each sample from Row A of the ligation stage plate to the corresponding wells of row A on the plates labeled P1, P2 and P3.
3. Seal each plate with adhesive film and place them in cooling chambers on ice until ready to add master mix.

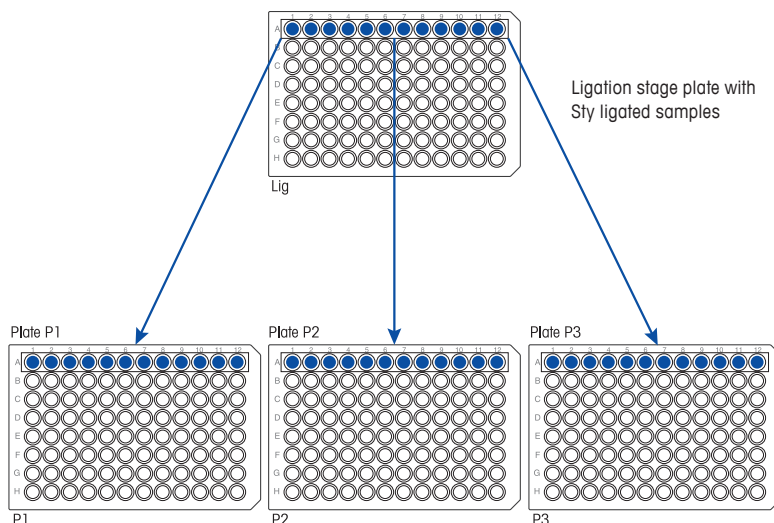


Figure 1: Transferring equal aliquots of diluted, ligated samples into three 96-well reaction plates.

An equal aliquot of each sample from the Ligation Plate is transferred to the corresponding well of each PCR plate. For example, an equal aliquot of each sample from row A on the Sty Ligation Plate is transferred to the corresponding wells of row A on PCR plates P1, P2 and P3.

### Adding PCR master mix to samples

Using Rainin's Liquidator 96, add 90  $\mu$ L of PCR master mix to each sample well of P1, P2 and P3. To avoid contamination, change the pipette tips after each dispense. The total volume in each well is now 100  $\mu$ L.

- Seal the reaction plates tightly with adhesive film.
- Vortex the center of each reaction plate at high speed for 3 seconds.
- Spin the plates for 30 seconds at 2,000 rpm.
- Keep the reaction plates in cooling chambers on ice until they're ready to be loaded onto the thermal cyclers.

With its ability to fill 96 wells at once, the Liquidator 96 has helped Affymetrix' genotyping team to improve the efficiency of their genotyping assays, saving valuable time and streamlining workflow. By switching their liquid handling devices from multichannel pipettes to the Liquidator 96, Affymetrix now saves as much as 30-45 minutes for every two PCR plates they prepare. Liquidator 96 streamlines many steps in PCR plate preparation, including adding master mix, sample dilution, sample distribution and sample pooling.



**"We use Rainin's Liquidator 96 for high-throughput applications. As it is versatile for many application areas, we replaced lower capacity pipettes with Liquidator 96 in PCR, purification and hybridization processes."**

Kay Mekar, Lab Supervisor, Scientific Services, Affymetrix Research Services Laboratory

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