

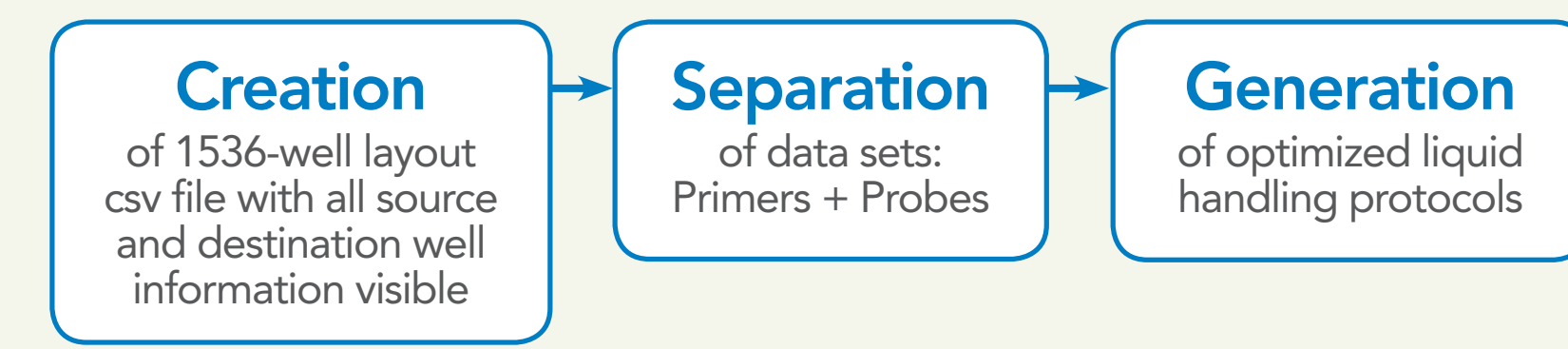
# Robotics Worklists for Creating 1536-Well Real Time PCR Assays

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## Overview

- New capabilities in the Nanobuilder software platform (used in conjunction with the Innovadyne Nanodrop™ or Nanodrop Express liquid handlers)
- Process vendor- or user-(LIMS) supplied plate map data sets in a csv file format.
- Csv files are presented in an easily visualized 1536 well-geometry format.
- Each of the 1536 destination wells contains all information about chemicals within the wells and their corresponding source plate and well.
- Nanobuilder software splits data into sample and primer information.
- Nanobuilder converts information into aspirate and dispense sequences optimized by time or plate lay-out.

- Different approach to worklisting for a liquid handler:
  - Traditional worklist approach: each aspiration/dispense step described in series of commands
  - Csv plate map worklist approach: describes desired end-result. Commands and actions delegated to liquid handler
- Many previously developed probe/primer combinations and the ability to modify or customize these for the user's needs.
- A test case of 48 DNA samples tested against 8 different probe/primer sets with 4 replicates of each reaction condition is presented.



## Introduction

As the field of genomics increasingly become higher throughput, the demand for lower volume assays and more effective automated control of these experiments increases. Several manufacturers have recently expanded their offerings of PCR plates to include 1536-well versions, however, the handling of large numbers of samples and primers creates a difficulty in programming automated liquid handlers to perform all the proper combinations of reactions in a logical order and with a logical plate layout, as well as with the desired number of experimental replicates.

- Genomics applications tend to be combinatorial in nature, e.g. M samples against N primer sets in X replicates. Some examples:
  - 4 primers against 128 samples in triplicate
  - 32 primers against 24 samples in duplicate
  - 1536 primers against one sample
  - 8 primers against 24 samples in triplicate plus 10 primers against 48 samples in duplicate (combining experiments)

- A logical set-up for these reactions can be easily comprehended for 384-well plates, but 1536-well plates can be difficult to comprehend the seemingly infinite number of data sets involved.
- This confusion may cause inefficiencies and errors in the assay development process.

In order to provide maximum flexibility for the user, it is desirable to utilize a worklist that the user can generate and modify to create custom plate layouts for higher density plates. Coupling this worklist to a software platform that is capable of translating the required plate map into actions by the liquid handler, as well as aiding in the visualization of this process for the user, results in an automation platform that is able to fully utilize the benefits of the lowered reagent costs and increased speed that higher density PCR platforms enable.

## About the Software and Instrumentation

- NanoBuilder Software control software developed by IDEX Health and Sciences, LLC for Innovadyne products
- Innovadyne Nanodrop II or Nanodrop Express low-volume liquid handlers are used to run the resulting protocols
  - non-contact, full aspirate and dispense
  - Unique architecture of the liquid handler eliminates reagent contact with moving parts
  - individually addressable solenoids (for dispensing) and syringe valves (for aspiration) to make such combinatorial protocols feasible
  - Motion control platforms include plate nest for dispensing in multiple plate orientations.



Nanodrop™ is a licensed trademark of IDEX Health & Science LLC.

## Workflow/Case Study

### 48 DNA samples tested against 8 different probe/primer sets with 4 replicates

- Source plates are in 96-well plate format
- A csv file is generated (see picture of partial file below):
  - Identify the most logical and/or quickest pattern with which to dispense each of the primers and samples
  - IDEX employs a tool that assists in setting up the csv file for this dispense pattern
  - Many LIMS systems would be capable of setting up the file as well

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