Customer name: GeneMark

Key user: Mark Walker

Application: The GeneMark lab is a relatively high throughput parentage lab based in Hamilton, New Zealand. The facility predominantly services the domestic bovine dairy industry and provides parentage determination from a single animal to a single parent all the way up to the entire crop of calves matched to both parents. Herd sizes average just over 500 animals and can be as large as several thousand.

Daily throughput in the laboratory is in excess of 3,000 animals per day during peak demand and most samples are processed using Sequenom’s iPLEX Platinum PCR chemistry and MassArray machines. The chemistry is the addition of PCR mastermix to magnetic bead extracted DNA followed by PCR cycling. At various times in the process, there are 3 additions of mastermix followed by PCR cycling. The first addition of 1.5 microlitres of mastermix is added to 1.0 microlitres of DNA in a 384-well PCR plate. The two subsequent mastermix additions are a further 1.0 microlitre each, added later in the process. The Tempest is currently responsible for the addition of each of the 1.0 microlitre mastermix additions.

Previous method: Accurate preparation of 10 or more 384-well plates per day is not something which is easily achieved manually, especially when the PCR volume is low. For this reason, the laboratory moved away from manual plate preparation (using single and multichannel hand-held pipettes) in favour of automation fairly early on. The previous automation used for the 1.0 microlitre addition was an Innovadyne Nanodrop II. It is a piezoelectric non-contact dispenser which specialises in low volume (microliter to sub-microlitre) dispensing.

User comments: The accuracy and speed with which the Innovadyne could deliver these volumes across a 384-well plate is well within our tolerances and was also the most suitable for our purposes at the time of purchase. However, it requires either an 8 place reservoir to hold liquid for each of the 8 channels or a reservoir trough. Even using small 0.2ml PCR tubes as a reservoir, the dead volume of PCR liquid wasted is quite high compared to the total volume dispensed.

The PCR mastermix is the most expensive part of the process so any reduction in dead volume wasted is a valuable saving. The Tempest is able to use either an external reservoir (like an Eppendorf tube or even a 15-50ml Falcon tube) of any size and recover all PCR mastermix within the lines for re-use. More importantly, it is able to use a single pipette tip as a reservoir for all 8 channels. The liquid path from this pipette tip reservoir through
the micro channels and the diaphragms in the Tempest chip is also very short, meaning little is wasted. This leads to a sizable reduction in the amount of mastermix the lab must prepare for a run of plates and as the throughput continues to grow, this saving grows along with it.

The Tempest is an exceedingly simple machine in comparison to the Innovadyne it replaces. Where the Innovadyne requires degassing of the system liquid with helium and this takes a considerable amount of time each day, the tempest (which utilises diagrams of fixed volumes) is ready to function immediately on start up. The diagrams have the bonus of being unaffected by liquid viscosity or ambient temperature and humidity, making them very accurate for small volumes where these factors have significant influence.

Both platforms are non-contact dispensers which deliver a 1.0 microlitre volume very rapidly across 384 wells. The tempest is very simple to program in terms of dispensing to some wells and leaving others blank, something which requires more involved programing in the machine it replaces.

The trade-off is that the volumes dispensed by the Tempest are less flexible because the total delivered must be devisable by the size of the diagrams each chip has to choose from. However, this is not a significant factor for the specific volumes we are working with and is a minor trade-off for the savings in time and waste reduction the Tempest provides.