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AUTOMATION OF A DNA IQ PURIFICATION METHOD FOR CASEWORK SAMPLES ON A TECAN FREEDOM EVO WITH FIXED TIPS.

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Our laboratory is currently developing an automated DNA purification protocol for casework samples on a Tecan Freedom EVO workstation equipped with fixed tips, using Promega DNA IQ™ magnetic beads system. A manual DNA IQ purification protocol has recently been validated and implemented into casework in our laboratory, and we are now proceeding with its automation.

The Freedom EVO workstation is equipped with a liquid handler arm (LiHa) with eight fixed tips and 2.5 mL syringes, a robotic manipulator arm (ROMA), an integrated plate sealer, a microplate centrifuge, a bar-code reader (sample tracking), a Tecan shaker, a magnet station and a heat block. The workstation is integrated with our in-house LIMS (www.DNAprofiles.ca), which generates worklists used by the TECAN Gemini software for automated preparation of purification plates. Versatile scripts minimize user intervention.

Key steps of the purification protocol are performed in closed/sealed containers to minimize the risk of contamination. The proteinase K lysis and filtration steps are performed manually, after which the lysate is stored in a bar-coded 1.5 mL micro-tube until processing on the workstation. At the purification step, a pierceable membrane cap is manually placed on the micro-tubes prior to their positioning on the workstation. Each 400 μ L lysate is transferred to its corresponding position in a 96-well deepwell (2,2 ml) plate with Promega lysis buffer (800 μ l) and magnetic beads. The plate is sealed with an easy-pierce foil seal. After incubation at room temperature, with shaking, the plate is transferred to the magnet station and the supernatant is discarded. To insure maximal recovery of magnetic beads and DNA during subsequent steps, Promega lysis buffer is added and the samples transferred to a 96-well microplate for washing and elution (5 minutes at 65°C on the heat block). At the final step on the magnet station the eluted DNA is pipetted into ABgene microtubes equipped with septa plugs and a unique 2D bar-code embedded underneath.

Our validation parameters include pipetting precision, optimization of magnetic beads recovery at all steps, and a thorough investigation of sample to sample contamination risks. The operation scripts are optimized to maximize throughput without compromising quality and reliability. DNA yield and quality are compared directly between manual DNA IQ purification and automated processing protocol.