

Automation of COVID-19 RNA purification using HALO™ Mag nucleic acid extraction kit on VERSA 10

I. Summary

The COVID-19 epidemic has placed a large burden on public and private testing laboratories. For every one positive case, 100's or 1000's of potential patient samples are screened. This rapid increase in the number of samples that need to be processed, as well the need for short turn around times, often outstrips current laboratory capacities. Further, the significant health risk that the Coronavirus presents to human health means that every safety precaution needs to be taken when handling potentially infected samples.

Aurora Biomed's experience automating pathogen detection workflows combined with the HALO™ Mag 100 nucleic acid extraction kit being a part of HALO 1 - Diagnostic kit for the detection of new SARS CoV-2 infections, provides rapid and reliable RNA isolation from patient samples (oropharyngeal or nasopharyngeal swab).

The optimized reagent chemistry ensures impurities are efficiently removed by a series of quick wash steps, yielding pure, high quality RNA ready for use in downstream analyses.

Once 100µL aliquots of sample has been transferred to a 96-well plate, Aurora Biomed's VERSA 10 automated liquid handling platform is capable of automating the entire nucleic acid extraction workflow, increasing sample throughput, while also freeing up staff to carry out more vital tasks. For the purpose of this validation, viral RNA was isolated from patients' samples (oropharyngeal or nasopharyngeal swab) stabilized with transport media.

II. Material and Methods

Equipment and virus RNA extraction kit

The HALO™ Mag 100 nucleic acid extraction kit (LAB8 Ltd., Belarus) has been optimized to isolate pure, high quality viral RNA/DNA from clinical samples. This kit allows for variable sample numbers (1-96 samples) to be processed at any given time and has been designed with automation of the workflow in mind.

The VERSA 10 automated liquid handling platform (Aurora Biomed Inc, Canada) is a flexible, open system that can be equipped with a 4- or 8- channel pipetting head that includes single-channel functionality (Figure 1).

The COVID-19 qPCR test kit A was used to evaluate the extraction of the viral RNA.

Automated workflow

The separation plate holding the 100µL patient samples is placed on the deck of the VERSA 10, from which point the entire RNA extraction workflow is automated. Paramagnetic beads addition, followed by lysis, RNA binding to the beads, wash steps, and elution of the purified RNA are performed as recommended by the HALO™ Mag 100 nucleic acid extraction kit user manual. The VERSA 10 deck configuration (Figure 2) and the HALO V automation protocol have been designed to maximize RNA yields and reduce the risk of cross-contamination.



Figure 1: VERSA 10 Gene Workstation

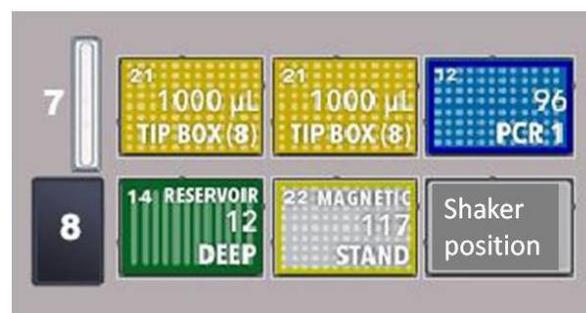


Figure 2: The deck layout of the VERSA 10 used to automate the HALO™ Mag 100 nucleic acid extraction kit workflow. Position #7 is the liquid waste disposal site, #8 is the tip disposal chute.

III. Results

The HALO™ Mag 100 nucleic acid extraction kit as a part of “HALO 1” - Diagnostic kit for the detection of SARS CoV-2 is designed with automation in mind and compliments the VERSA 10's powerful automation capabilities to reliably purify viral RNA.

To validate the automation of the HALO™ Mag 100 nucleic acid extraction kit, viral RNA was isolated from 48 patient samples. The viral RNA isolated from 48 putative patient samples was then analysed using a commercially available qPCR-based COVID-19 test kit.

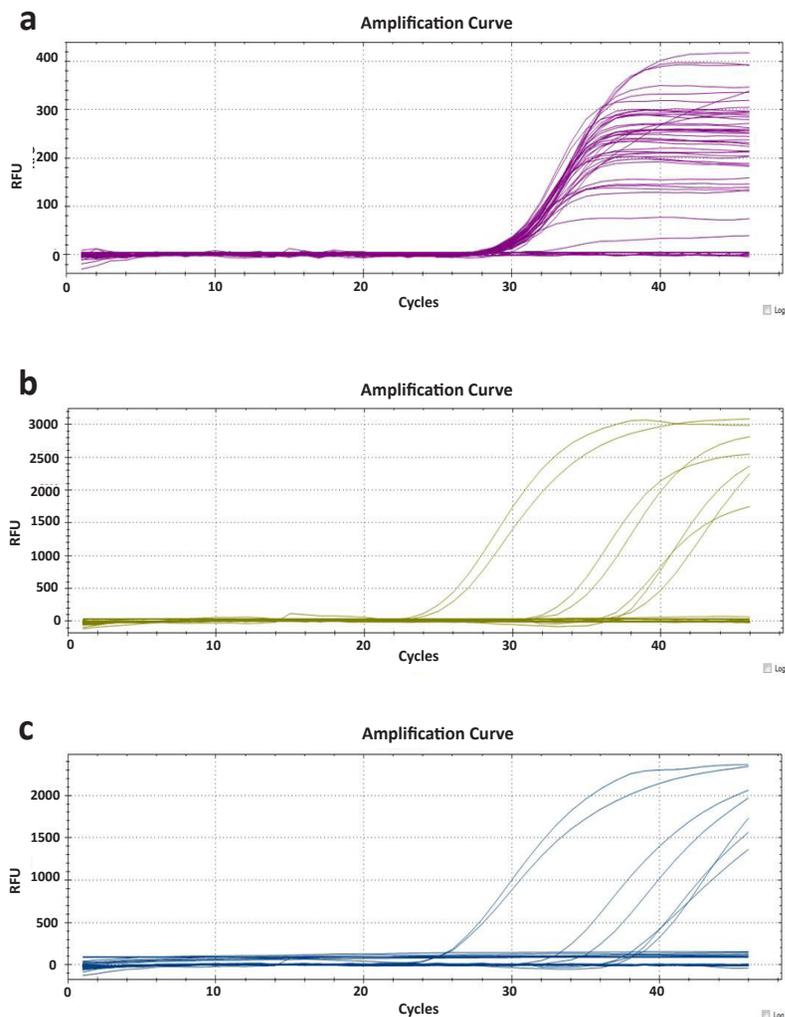


Figure 3: Test kit A results for reaction 1 performed on 48 samples extracted using the HALO™ Mag 100 nucleic acid extraction kit. Seven positive samples were identified. (a) amplification curves for internal control, (b) amplification curves for ORF 1ab gene, (c) amplification curves for N gene.

qPCR test kit A is comprised of two separate qPCR reactions per sample, both with the internal control. The first reaction tests for the presence of COVID-19 ORF1ab and N genes, the second for the presence of the E gene. Test kit A has a positive test result Ct detection threshold of <38 and negative result detection threshold of >40.

Test kit A identified the seven of 48 samples as being positive for COVID-19. The E gene probe set detected the E gene with a Ct of >40 for two additional samples.

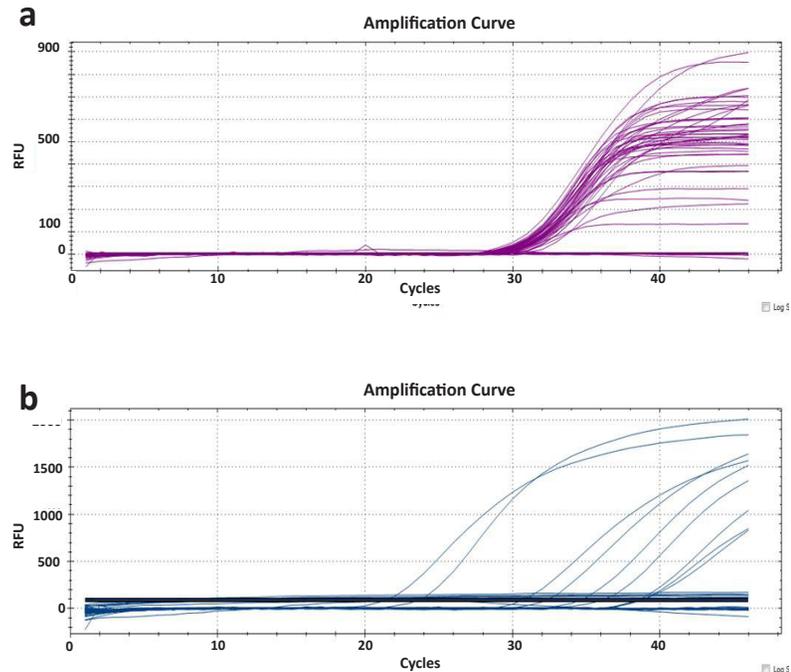


Figure 4: Test kit A results for reaction 2 performed on 48 samples extracted using the HALO™ Mag 100 nucleic acid extraction kit. Seven positive samples were identified. (a) amplification curves for internal control, (b) amplification curves for E gene.

IV. Conclusion

Combining the power of HALO™ Mag 100 nucleic acid extraction kit with the precision of Aurora Biomed's VERSA 10 automated liquid handling platform provides compact solution for the isolation of pure, high quality viral RNA from patient samples. Moreover, the instrument minimizes the risk of manual errors and cross-contamination.

The VERSA 10 increases reproducibility and the number of samples that can be processed at any given time. VERSA 10's flexibility allows users to automate RNA extraction as well as PCR setup, sample normalization and general liquid handling applications on the same workstation.