# Are Point-of-Care devices suitable for the management of Dengue Virus infections? A comparison with mainframe analysers using External Quality Assurance (EQA) data

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## Introduction

Dengue virus (DV) is an arthropod-borne disease that has a significant impact on human health and the economy[1]. Mosquitos are the primary transmitters of this viral infection. Globally, around 2.5 billion individuals have been affected by the virus[2]. Early and rapid dengue identification is crucial for monitoring transmission dynamics and enabling swift responses to outbreaks[3].

Access to pathology testing remains limited in many countries due to geographical distances and a lack of facilities and resources. Traditionally, potentially cost-prohibitive enzyme-linked immunosorbent assays (ELISAs) and Fluorescent immunoassays (FIA) on mainframe analysers are used to detect DV. Point of Care (POC) devices are an accessible resource and can be performed in resource-limited settings[4].

The RCPAQAP has a Dengue Virus Serology EQA program covering DV IgG, IgM and NS1 Ag markers suitable for both mainframe and POC assays.

We sought to evaluate the diagnostic accuracy of POC devices compared to mainframe ELISA/FIA assays based on the returned EQA results.

## Method

Clinical serum/plasma samples from known positive/negative DV patients were sourced from Australian and overseas laboratories. Additional NS1 Antigen (NS1 Ag) positive samples were produced by spiking NS1 Ag (type 4) inactivated cell culture fluid into negative serum samples. The samples were further tested using automated ELISA methods to confirm the expected positive/negative results prior to dispatch.

A total of 20 samples (4 per year) representing the NS1/IgM/IgG disease phases (as shown in Figure 1) were distributed to an average of 110 participating laboratories over a five-year study period (2019-2023). The acute phase samples consisted of 4 spiked NS1 Ag and 1 native NS1. There were also 6 IgM/convalescent, 2 IgG/recovered, and 7 confirmed negative samples.

Returned quantitative and qualitative (positive/negative/equivocal) results from mainframe analysers were reviewed and compared to the qualitative (positive/negative) POC results using RCPAQAP in-house software.

## Results

Mainframe analysers returned >80% positive consensus for each of the 5 NS1/acute phase samples compared to an average of 40% for the POC devices. One of the IgM/convalescent samples achieved 94% and 78% positivity for mainframe and POC, respectively; the other was reflective of a lower IgM at 56 and 5%. The 2 IgG/recovered specimens returned ≥90% positivity for IgG for all mainframe methods, and 53% for the POC methods (Table 1, Figure 2). As shown in Table 1, combining positivity for any of the analytes achieves improved sensitivity for POC devices that now measure all three[5].

There was >95% consensus for all methods for each of the negative samples.

## Discussion

Based on this study, overall, the mainframe methods performed as expected for all 20 samples.

POC methods were less sensitive for detecting NS1 in the acute samples. Similarly, for IgM in the convalescent samples and IgG in the recovered samples. However, consistent with previous studies, improved sensitivity by combining positivity for any of NS1/IgM or IgG reflecting the cross-over of levels at different phases (Figure 1, Table 1).

The >95% agreement for the true negatives confirms an acceptable specificity for all methods.

Overall, POC devices demonstrate acceptable performance compared to mainframe analysers.
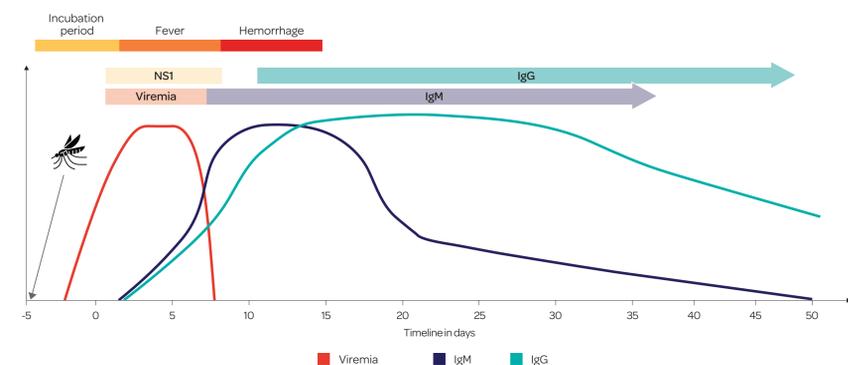


**Figure 1**    Immune response to a DV infection showing the associated disease phases.[4]
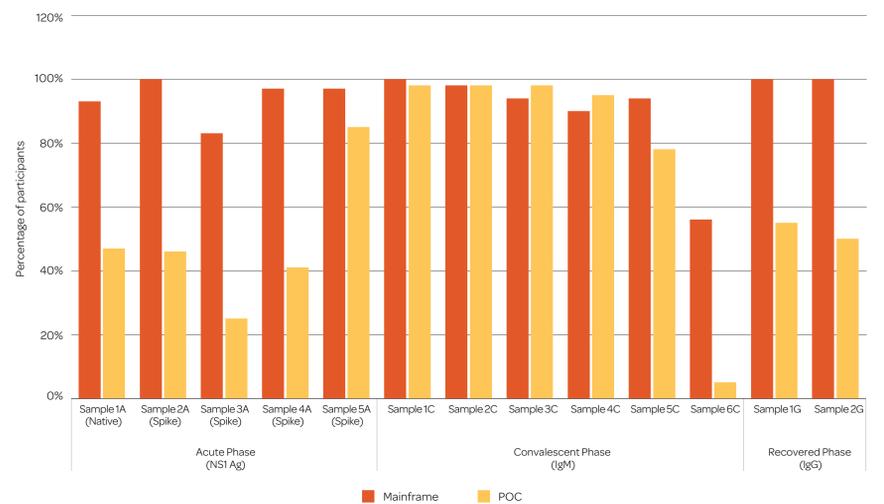


**Figure 2**    Percentage of reported positive interpretations for the acute, convalescent and recovered phase samples.

**Table 1**    Comparison of positivity rates for the individual phases vs a combination of all three phases.

| Phase | Sample | NS1 | | IgM | | IgG | | Combined Sensitivity of >80% for NS1/IgM/IgG | |
|---|---|---|---|---|---|---|---|---|---|
| | | Mainframe (%) | POC (%) | Mainframe (%) | POC (%) | Mainframe (%) | POC (%) | Mainframe (%) | POC (%) |
| Acute (NS1) | 1A (Native) | 93 | 47 | 100 | 100 | 100 | 90 | ✓ | ✓ |
| | 2A (Spiked) | 100 | 46 | 0 | 2 | 0 | 0 | ✓ | ✗ |
| | 3A (Spiked) | 83 | 25 | 0 | 1 | 0 | 0 | ✓ | ✗ |
| | 4A (Spiked) | 97 | 41 | 0 | 2 | 0 | 0 | ✓ | ✗ |
| | 5A (Spiked) | 97 | 85 | 2 | 1 | 0 | 2 | ✓ | ✗ |
| Convalescent (IgM) | 1C | 0 | 0 | 100 | 98 | 100 | 99 | ✓ | ✓ |
| | 2C | 0 | 0 | 94 | 98 | 94 | 98 | ✓ | ✓ |
| | 3C | 0 | 1 | 90 | 95 | 100 | 99 | ✓ | ✓ |
| | 4C | 0 | 0 | 98 | 98 | 90 | 99 | ✓ | ✓ |
| | 5C | 0 | 0 | 94 | 78 | 0 | 0 | ✓ | ✗ |
| | 6C | 0 | 2 | 56 | 5 | 90 | 97 | ✓ | ✓ |
| Recovered (IgG) | 1G | 0 | 0 | 17 | 1 | 100 | 55 | ✓ | ✗ |
| | 2G | 0 | 9 | 0 | 2 | 100 | 50 | ✓ | ✗ |

## Conclusion

Based on the returned EQA results in this study POC devices are suitable for use where mainframe analysers are not readily available, however awareness of variation in sensitivity and specificity between methods for patients who present in different stages of their infection is important when interpreting results.

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