

affigene® CMV trender in QCMD proficiency program 2008

Background

Quality Control for Molecular Diagnostics (QCMD) specialises in the standardisation and quality control for molecular diagnostics and genomic technologies. A proficiency panel for Cytomegalovirus (CMV) was sent out in May 2008 to 188 participants from 32 different countries. 216 datasets were reported to QCMD of which 67 were analysed with commercial real-time PCR assays. Three datasets were analysed using affigene® CMV trender, one of which submitted by Cepheid. This report presents the result for the Cepheid affigene® CMV trender dataset.

Material and methods

The QCMD CMV panel 2008 consisted of ten samples, with varying CMV viral concentration. The concentration was unknown to the operator at the time of analysis. The QCMD panel was prepared using affigene® DNA extraction. Subsequently, the samples were analysed by real-time PCR using affigene® CMV trender on the Mx3000P instrument (Stratagene, La Jolla, CA). Each laboratory reported their results directly to the QCMD office.

Results and discussion

Of the ten blinded samples, two was expected to be negative CMV samples. The other samples had expected viral loads ranging from 200 copies/ml to 2×10^6 copies/ml.

Qualitative results

QCMD used a scoring system ranging from 0 points ('highly satisfactory') to 3 ('highly unsatisfactory'). A correct result for the qualitative performance of the panel gave 0 points. Depending on the sample concentration, an incorrect results gave between 1 and 3 points.

The affigene® CMV trender data set achieved the best possible scoring of 0 points, with all ten samples correctly determined as positive or negative respectively.

For the reported 67 data sets which were analysed with commercial real-time PCR assays, the mean scoring was 0.4 (Table 1). Of all reported qualitative datasets, 74.1% detected all samples.

Average value commercial real-time PCR (n=67)	Average value affigene® CMV trender (n=1)	Maximum score attainable
0.4	0	0

Table 1 Qualitative results for commercial real-time PCR assays.

A correct result gives 0 points. Depending on the sample concentration, incorrect results give 1-3 points. The best obtainable score is 0. The table shows the average values for the 67 commercial real-time PCR datasets, and the value for the affigene® CMV trender dataset submitted by Cepheid.

Quantitative results

Figure 1 displays the quantitative results of the CMV proficiency panel. The viral load is plotted versus QCMD sample number. The figure shows the results from all 66 data sets analysed with commercial real-time PCR assays that reported quantitative results. The reported mean for the commercial assays is displayed for each panel member, as well as the standard deviation. The expected viral load and the result from the Cepheid affigene® CMV tender data set are showed separately.

The results for all samples from the affigene® CMV tender data set were within one standard deviation of the reported mean. All samples received the maximum scoring also for the quantitative results. This score was only obtained by 21.5% of all reported quantitative datasets.

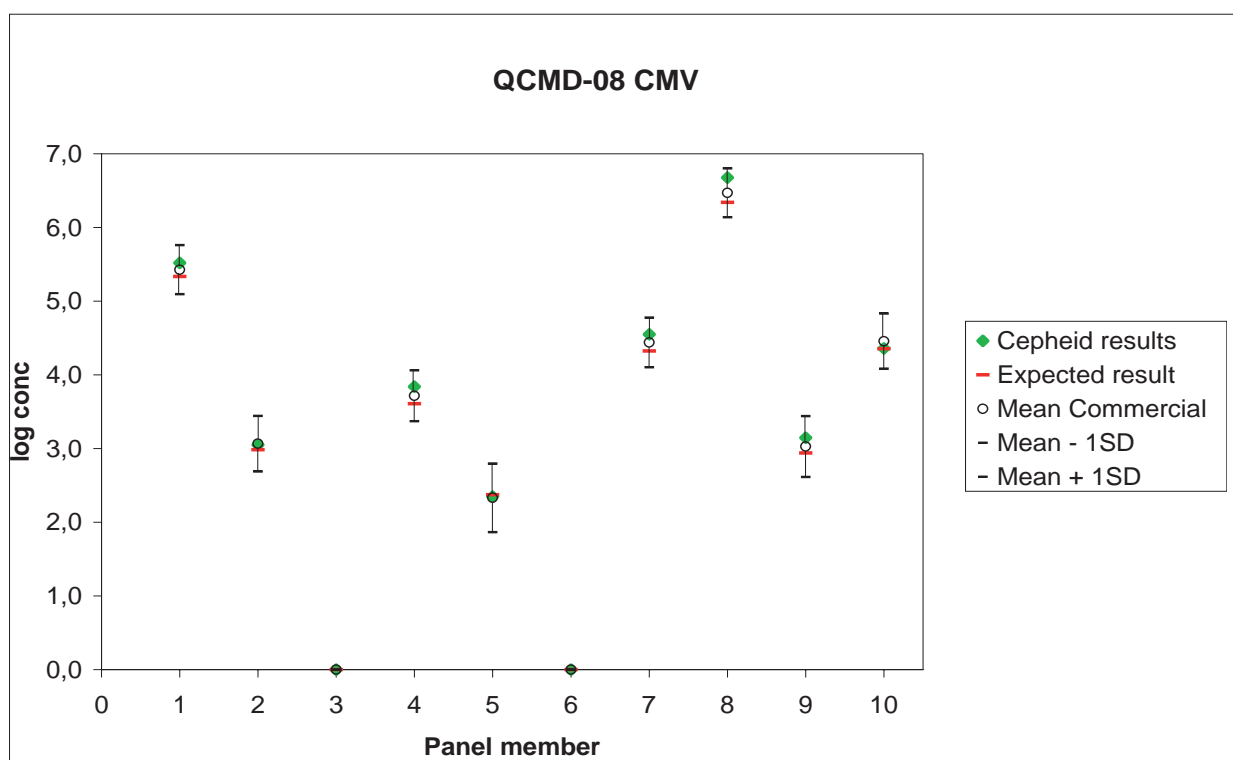


Figure 1. Results of the 2008 QCMD CMV panel.

The expected viral load for each panel member is displayed in red. The affigene® CMV tender data set submitted by Cepheid is displayed in green. The reported mean viral load from 66 data sets analysed with commercial real-time PCR assays is shown as an open circle(o), with bars indicating the standard deviations for each panel member.

Conclusion

- The affigene® CMV tender assay performed well in the QCMD CMV proficiency panel 2008.
- The assay shows excellent performance in the qualitative analysis.
- The quantitative results are all within one standard deviation from the average of all reported data.