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ePoster Viewing

Molecular virology

CMV, EBV and HHV6 viral load evaluation by the automated VERSANT k-PCR Molecular System in plasma and cerebral spinal fluid

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Objective. Quickness of results, sensibility and multi-specimen processing represent a major challenge for the molecular routine laboratory; this is especially important for viral load in immunocompromised patients and in subjects where a rapid diagnosis is essential to start a proper therapy.

In this study we try to establish the analytical performance of the Automated VERSANT kPCR Molecular System the for CMV, EBV and HHV6 load quantification in plasma and in cerebral spinal fluid (CSF) specimens.

Methods. Forty-three plasma and 11 CSF samples randomly collected from patients serially admitted to University Hospital Policlinico Umberto I of Rome were examined.

The VERSANT kPCR Molecular System with new VERSANT MiPLX Software Solution (Siemens Healthcare Diagnostics, Tarrytown, NY, USA) was used to extract DNA from samples. CMV, EBV and HHV6 amplification, detection and quantification were performed automatically on the AD module of the VERSANT kPCR Molecular System, adding quantitative standards in each run. Data obtained were compared with those from a currently used semi-automated system including EasyMAG (Biomerieux, Italia S.p.A.) extraction and Elite-MGB (ELITech Group S.p.A. Molecular Diagnostics, Italia) Real time PCR Amplification.

Results. The VERSANT kPCR Molecular System with new VERSANT MiPLX Software Solution allowed to simultaneously process 43 plasma samples in less than 3 hours.

In plasma samples only EBV and CMV load was analyzed. Detectable levels of CMV DNA were detected in 24 out of 43 samples and a good correlation was observed by data obtained from the two assays used ($r=0.96$ $p<0.0001$). Three samples showed undetectable levels of CMV DNA using both methods; 16 specimens showed viremia values detectable but <250 copies/ml using ELITech System; 7 of these were quantified using VERSANT kPCR while in the remaining samples the viremia was undetectable by using both assays. As regard EBV load, 39 out of 43 plasma samples were undetectable by using both systems. EBV DNA was quantified in 4 samples: in 2 of them, EBV was detected by both methods, in 1 only by VERSANT kPCR and the in the other one by ELITech System.

As regard CSF samples, CMV DNA was quantified in 2 samples of which 1 was detected only by VERSANT kPCR. EBV DNA was quantified in 4 samples: in 2 of them, EBV was detected by using both methods; in 2 EBV was detected only by ELITech System. No difference was observed in the detection of HHV6 (2 positive out of 11 examined). VERSANT kPCR also allowed to discriminate the two HHV6 variants A and B.

Conclusions. The new VERSANT MiPLX Software Solution proved to be flexible in his ability to combine extraction and amplification. In plasma samples, a good correlation was observed when data are compared to those obtained by ELITech system.