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P0818 Validation of a fast real-time PCR assay for herpes simplex virus 1/2 and varicella zoster virus detection directly from cerebrospinal fluid

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Background: Viral and bacterial meningitis cannot be differentiated without analysis of cerebrospinal fluid (CSF), done primarily to exclude bacterial meningitis. Identification of the specific virus also informs on prognosis, enhances patient care, reduces the use of antibiotics, decreases length of stay in hospital, and may prevent spread of infection. Moreover, the severity of herpetic meningitis/encephalitis is a special case, to be treated as an emergency.

We aimed to evaluate a new, fast, multiplex real-time PCR assay (Eurobioplex EBX-030 alpha herpes virus CSF direct - Eurobio-Scientific, Les Ulis, France) for the simultaneous detection of HSV1, HSV2 and VZV, directly from CSF without DNA extraction.

Materials/methods: First, an assay lasting 1h 20 minutes for the detection of HSV1-HSV2-VZV directly from CSF, was evaluated on 16 HSV/VZV negative CSF, spiked with quality control molecular diagnostic of HSV1–HSV2-VZV (QCMDs, Qnostics), and on 2 HSV1 positive samples.

Second, EBX-030 alpha herpes virus CSF direct, a new faster assay lasting 40 minutes, was evaluated on serial dilutions of an HSV1 positive CSF, spiked with HSV2 and VZV QCMDs, and on 17 clinical CSF samples, positive or negative for HSV1, HSV-2 or VZV. An internal control was included for both assays.

Results: Using the longer assay, Ct values for equal final amounts of viral DNA were similar on extracted DNA or directly on CSF. For instance, HSV1 was detected with a Ct=38.4 (n=7) directly on CSF, versus a Ct=38.3 (n=4) on extracted DNA.

Analytical sensitivity was as good with the shorter PCR program, and around 10 copies/microliter for the 3 viruses. Time from sample to result, was reduced by half, to 40 minutes, which constitutes a real advantage for emergency use.

With the faster assay EBX-030, sensitivity and specificity on clinical samples were 100 % for the 3 viruses. A larger sample size is under study.

Conclusions: While the new fast real-time PCR for detection of HSV1, HSV2 and VZV EBX-030 can still be used with extracted DNA from any tissues or fluid, it shows excellent performance and provides a fast and convenient diagnostic tool to monitor viral meningitis directly from CSF.