REAL-TIME PCR STANDARIZATION PANEL FOR ENCEPHALITIS DIAGNOSTIC IN THE LIGHTCYCLER NANO (TM) ROCHE, WHICH INCLUDE DNA AND RNA VIRUSES

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Introduction: Encephalitis is an inflammation of the brain structures: neurons, vessels or glia cells. Encephalitis is an important cause of morbidity and mortality worldwide especially in children. However, both the incidence and case fatality are thought to be underestimated and underdiagnosed. If infection is not fatal, individuals often have severe physical, cognitive, emotional, behavioral, and social difficulties. In the last years statistics showed that viruses are the most common pathogens that cause this disease including both DNA and RNA viruses. Nowadays Real- Time PCR is a very useful assay to detect and diagnose a lot of infections whit a high specificity and sensibility. A panel of real- time PCR assays based on *Taq*Man technology has been developed for the detection of 5 different viruses. The ability to use uniform PCR conditions for all assays permits simultaneous processing and detection of many different viruses, thus economizing the diagnostic work.
Materials and Methods: Positive controls from Vircell Herpes Simplex Virus 1(HSV), Cytomegalovirus (CMV) were used and positive samples of Varicella Zoster Virus (VZV), Epstein Barr Virus (EBV), and Enterovirus acquired from state laboratory of Public Health (LESP, Guadalajara). All virus tests were designed to be conducted under identical cycling conditions, to facilitate the molecular diagnostic work. The assay was developed in LightCycler Nano (Roche). Before the real- time PCR we corroborated the specificity of all the primers and probes in an end point assay. The gel was runto check only one band after electrophoresis.
Results: After several assays to found the best conditions for the real- time PCR we could developed a panel for 5 different viruses with se same conditions for all of them. The assay was standardized qualitative and quantitative to determine load viral.
Conclusion: This panel will provide us a high specificity and sensibility assay to detect 5 different virus at the same time. Whit this we would save time for the diagnostic work.