利用即時聚合鏈鎖反應評估母奶中人類巨細胞病毒之研究 Evaluation of Human Cytomegalovirus in Breast milk by Using Quantitative Real-Time Polymerase Chain Reaction Assay

中文摘要

爲了新生兒的健康，近年來政府積極鼓勵哺育母乳。特別是早產兒，可由母乳的哺育得到多方面的好處。然而在亞洲地區，育齡婦女的人類巨細胞病毒 (HCMV) 感染率相當高。根據研究發現，母奶是嬰兒期得到 HCMV 感染的主要來源之一。對於免疫力不全的早產兒而言，在出生後經母奶得到 HCMV 感染時，可能會發生嚴重的併發症或導致神經發展上之後遺症。目前臨床評估 HCMV 感染的檢驗方法，既耗時且未能反映出真實臨床意義。而真正導致病毒感染者，卻與病毒量相關。因此，本研究藉由利用即時聚合鏈鎖反應，偵測經母奶中人類巨細胞病毒之病毒量，希望能早期精確評估經母奶傳染人類巨細胞病毒給早產兒與病毒量之相關性。凡出生體重小於 1500 百公克以下；週數小於 35 週之早產兒均被收案。早產兒於出生三週內、第四、八及十二週各留小便，作病毒培養。母親生產後五天內、第二、四、八及十二週，各收集母奶檢體保存在攝氏 -70oC 之冰箱中，進行即時聚合鏈鎖反應定量母奶中 HCMV 量。研究結果顯示母乳檢體中，早期有較高人類巨細胞病毒病毒量 (1200.22 ± 180.23 copies/ml)，3 位早產兒在出生後平均 65.33 天被病毒感染。被人類巨細胞病毒感染的早產兒臨床上會出現，如：類敗血症狀、顆粒球低下、血小板數偏低及延遲性黃疸等症狀。由研究証實，即時聚合鏈鎖反應，是一準確、快捷、方便、及低污染，能應用在評估經母奶傳染人類巨細胞病毒給早產兒疾病監測及診斷上。母奶早期出現較高人類巨細胞病毒量與早產兒被感染是有相關，爲病毒感染的危險因子。因此，建議母親之人類巨細胞病毒血清抗體呈陽性，且早期母奶有較高病毒量者，餵食早產兒之母奶應先進行冷凍處理後才可餵食。

英文摘要

For the health of infants, especially preterm infants, the government has been promoting breast-feeding for many years. Seropositive rate for human cytomegalovirus (HCMV) in pregnant women was very high in Asia. According to previous studies, the shedding of the virus into breast milk was the main source of transmitting HCMV to their tiny babies. However, postnatal CMV infection in preterm infants might cause serious complications and neurologic sequelae of life. There were many clinical methods to detect HCMV, however, these techniques were time consumption, too sensitive to be clinically useful. In fact, viral load might correlate with HCMV transmission. We, therefore, assessed viral load in human milk for transmission of HCMV to preterm infant using quantitative real-time PCR assay.

This prospective study comprised all breastfed preterm infants with a birth weight of less than 1,500 g and a gestational age of less than 35 weeks, who hospitalized in the neonatal unit. Urine from baby for HCMV culture was obtained within 3 weeks after birth, and at the 4th, 8th and 12th week. Breast milk was collected within day 5, and at the 2nd, 4th, 8th and 12th week for in vitro analysis. All the breast milk samples were frozen at – 70°C until they were screened for CMV infection by using qRT-PCR assay. Based on the result of our study, 3 preterm infants were infected at mean of 65.33 days after birth. The high HCMV viral load was detected earlier in breast milk samples of transmitters (1200.22 ± 180.23 copies/ml). Sepsis-like symptoms, neutropenia, thrombocytopenia and prolonged jaundice, were found in these infected infants. In conclusion, quantitative real-time PCR is a precise, rapid, convenient, and low contamination diagnostic method to assess human cytomegalovirus load in breast milk. The high viral load detected earlier in breast milk, which might correlate with HCMV transmission. This was a risk factor for viral transmission to preterm infant. Therefore, we suggest that inactivation of the virus in breast milk from seropositive mother by freezing may be a way of reducing the transmission of this virus via breast milk to preterm infant.