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Diagnosis of Chlamydia Infection in Endocervical Swabs by Real Time RT-PCR

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Background & Objectives: Infection of reproductive organs with Chlamydia trachomatis may lead to severe complications such as salpingitis, ectopic pregnancy, premature rupture of membrane (PROM), peritoneal inflammatory disease (PID) and infertility. Effective screening for the infection can facilitate prompt treatments and prevent its sequelae. In this study, we evaluate the efficacy of diagnosis of the Chlamydia infections by quantitative real-time PCR.

Method: Endocervical swabs from symptomatic patients (n=89) were taken by several rotating two sterile cotton sticks, which one was for the RNA amplification (RNAamp) assay [VIDAS Chlamydia kit (bioMerieux, Lyon, France)], and another for the quantitative rael-time PCR (qPCR) assay [RealArt™ C. Trachomatis PCR kit (Abbott Diagnostics, IL, USA)]. The RNAamp and qPCR assay was performed by the manufacturer's protocol, respectively. For the qPCR, DNA was extracted and purified with AccuPrep DNA extraction kit (Bioneer, Daejeon, Korea). The quality and quantity of purified DNA was evaluated by gel electophoresis. The RealArt™ kit is a ready-to-use system for the detection of Chlamydia specific DNA for the major outer membrane protein with sequence specific primers and a dual labeled probe. Quantitative analysis of Chlamydia DNA is conducted with the standard curve assay using positive control samples in the kit. The clinical symptom, treatment and outcome were verified with documented records.

Results: The purified DNA was suitable for qPCR in 98.9% (88/89) of the endometrial swabs. The prevalence of the infection was 12.4% (11/89) by the RNAamp and 10.2% (9/88) by the qPCR. There were two cases of diagnostic discrepancy to the Chlamydia infection by the RNAamp and the qPCR assay. Titer range of qPCR in positive samples was from 30 to 4,500 copies of Chlamydia DNA per ml. All positive samples were confirmed by repeated qPCR assay, and the coefficient of variances between first and second qPCR were less than 10%. The infected patients (n=9) by the qPCR showed 3 of PID, 3 of vaginitis, 2 of PROM and 1 of intrauterine fetal growth retardation.

Conclusions: The RealArt™ C. Trachomatis PCR kit is very efficient for the screening of Chlamydial infections in endocervical swabs. Also, the quantitative data may be valuable for the confirmation of adequate treatments. We also found the adverse effect of Chlamydial infection on the outcome of pregnancy. Our results suggest that the diagnosis of Chlamydia infection should be systemically checked for successful outcome of on-going pregnancy and ART program.