

## Evaluation of Immunohistochemistry and PCR in Diagnosis of Toxoplasma Infection in Tissues of Human Aborted Fetuses

Bahador Sarkari,<sup>\*1</sup> Qasem Asgari,<sup>2</sup> Saeed Mirzaee<sup>2</sup>

Received: 27 Dec 2012

Accepted: 20 Jan 2013

Available online: 27 Feb 2013

1. Basic Sciences in Infectious Diseases Research Center, Shiraz University of Medical Sciences, Shiraz, Iran
2. Department of Parasitology and Mycology, Shiraz University of Medical Sciences, Shiraz, Iran

**T***oxoplasma gondii* is one of the most common protozoa that infect humans and a wide range of mammals and birds [1]. The infection is particularly important in women when they acquire the infection for the first time during their pregnancy where an intrauterine transmission of the parasite may occur. Effective prenatal diagnosis of congenital toxoplasmosis can permit a decision to terminate the pregnancy at the early stage or initiate the treatment of late-term fetus in uterus. Prenatal diagnosis is commonly performed based on biological and serological tests on fetal blood and amniotic fluid, and ultrasonographic examination of fetus [2]. The current study describes the performance of immunohistochemistry (IHC) and PCR methods in diagnosis of Toxoplasma infection in formalin-fixed paraffin embedded tissues of human spontaneous aborted fetuses. Paraffin-embedded blocks of aborted placenta which were collected during 2005 to 2009, from two university-affiliated hospitals in Shiraz were used in this study. DNA was extracted from the samples and a semi-nested PCR was used to amplify the segments of the 35-fold repetitive DNA region of B1 gene of *T. gondii*. The forward; 5'-GAAGTGCATCCGTTTCATGAG-3' and reverse; 5'-TCTTTAAAGCGTTTCGTGGTC-3' primers were used to amplify the related target. Cycling conditions for both the direct and semi-nested PCR were denaturation at 94°C for 5 min, followed by 40 cycles of 94°C for 1 min, 60°C for 1 min, and 72°C for 2 min.

For IHC, fifty paraffin-embedded blocks of aborted placenta which were found to be positive for *Toxoplasma* infection by PCR along with 50 samples of aborted placenta which were negative by PCR for Toxoplasma infection were evaluated by IHC.

Samples from liver of Toxoplasma infected mice were used as positive control while tissue samples from normal mice were used as negative controls.

No Toxoplasma cyst or tachyzoites were detected in any of PCR-positive samples. Moreover none of PCR negative samples were positive by IHC. Accordingly no agreement was found between PCR positivity for Toxoplasma and the results of IHC. When the tissue samples from Toxoplasma infected mice were tested by IHC, tachyzoites of parasite were detected in the tissues while no Toxoplasma cyst or tachyzoite were detectable in tissues of normal mice.

Previous studies showed that the prenatal diagnosis of congenital *T. gondii* infection, based on a PCR test performed on amniotic fluid is rapid, safe, and accurate approach [3].

In our study no parasite cyst or tachyzoites were detected in PCR positive sample by IHC. This might be contributed to the low level of parasite in the aborted tissues. Moreover, PCR is a very sensitive method and can detect a few copy of DNA in the samples while in IHC the whole parasite or tissue cyst must be presented to be detected by immunoenzymatic approaches. Considering the result of this study, IHC might not be a good approach for diagnosis of toxoplasmosis in congenital infection.

Corresponding author at: [sarkarib@sums.ac.ir](mailto:sarkarib@sums.ac.ir)

© 2013 Zahedan University of Medical Sciences. All rights reserved.

### References

1. Montoya JG, Liesenfeld O. Toxoplasmosis. Lancet 2004; 363(9425): 1965-76.
2. Kaye A. Toxoplasmosis: Diagnosis, treatment, and prevention in congenitally exposed infants. J Pediatr Health Care 2011; 25(6): 355-64.
3. Hohlfield P, Daffos F, Costa JM, et al. Prenatal diagnosis of congenital toxoplasmosis with a polymerase-chain-reaction test on amniotic fluid. N Engl J Med 1994; 331(11): 695-9.