Asian Journal of Water, Environment and Pollution, Vol. 19, No. 3 (2022), pp. 75-78.
DOI 10.3233/AJW220042

The Effect of Toxoplasmosis on Pregnant Women and Its Diagnosis by "VIDAS Test of IgG Avidity"

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Received July 17, 2021; revised and accepted September 17, 2021

Abstract: Toxoplasmosis is well known as a cause of infection in pregnant women. Although many serological methods are available, diagnosis of early Toxoplasmosis may be extremely difficult. Toxoplasmosis is typically diagnosed during pregnancy via testing of maternal serum for IgM and IgG anti-Toxoplasma antibodies. Toxoplasmosis is normally asymptomatic, but it can have serious effects in immune-deficient individuals. Because avidity increases over time during infection, determining specific IgG avidity allows for more precise dating. Long-term IgM persistence makes it difficult to distinguish between acute and chronic infection. Seventy-six women were tested for VIDAS lgM, lgG antibodies, and VIDAS toxo-IgG avidity during the first 16 weeks of pregnancy. Low avidity antibodies were found in two (33.3%) of the six IgM-positive sera and four (11.11%) of the IgG-positive sera. Low avidity was noticed in 2 (3.27%) of the 61 sera that were negative for IgM. The low avidity indicates a recent infection, whereas high avidity in 4 (50%) of the 6 positive IgM and 24 (74.74%) of the 33 positive IgG indicates a long-ago infection. These conclusions emphasise the importance of using "VIDAS IgG avidity" in conjunction with the VIDAS IgM and IgG assays to offer to prove the presence of acute infection from a single serum sample for pregnant women.

Key words: Toxoplasmosis, VIDAS test, avidity, early pregnancy**.**

Introduction

Toxoplasma gondii is one of the most common infectious agents in humans, with a global distribution. Acute infection is usually asymptomatic. However it was assumed that congenital toxoplasmosis was caused by an initial infection received during pregnancy (Berredjem et al., 2017; Thiebaut et al., 2007), but not by the reactivation of latent infection in immune-competent pregnant women (Rahbari et al., 2012). Whereas (Petersen et al., 2001) believed that latent Toxoplasma could reactivate and cause a congenital transmission of the parasite to fetuses. The most common method for diagnosing toxoplasmosis is the detection of *Toxoplasma gondii* specific antibodies. Unfortunately, classic serological approaches are usually not useful for distinguishing recent from past toxoplasmosis. Several investigations have confirmed the ability of particular IgM to remain for a long time, even at high levels (Ramzan et al., 2009). IgG avidity tests have been shown to provide confirmatory evidence of acute infection and they can distinguish reactivation from primary infections with a single serum specimen for pregnant and immunosuppressed patients (Kamani et al., 2010; Kompalic-Cristo et al., 2007). Thus, IgG produced in the first few months after primary infection has low avidity, whereas IgG produced months or years later has high avidity (Gutierrez-Zufiaurre et al., 2004; Pomares and Montoya, 2016).

A high level of IgG avidity results of the test using the newly developed VIDAS procedure is commercially available in Europe and has been reported to rule out

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an infection that happened during the previous four months, increasing the utility of avidity testing to the pregnancy's fourth month (Foroutan-Rad et al., 2016).

The aim of this study is to determine IgG avidity's performance method for the detection of anti-Toxoplasma antibodies in women who are pregnant throughout the first 16 weeks of pregnancy.

Material and Methods

Study Group

A total of 78 women in their first 16 weeks of pregnancy had their sera examined. The sera were submitted to outpatient clinics. The assay was carried out at the Laboratory in "Children's & Women's Hospital "in Al-Ramadi city.

VIDAS Toxoplasma IgM and IgG Assay

Serum and plasma were used for IgG and VIDAS IgM tests, which are quantitative tests performed by the ELFA mechanism, "Enzyme linked fluorescent assay". The assay uses a Sandwich approach using a two-steps enzyme immunoassay with fluorescence detection at the end. For the test, the solid phase receptacle (SPR) serves as the solid phase as well as the pipetting device. Reagents for the assay are ready to use and predispensed in the sealed reagent strips. All of the assay steps are performed automatically by the instrument after a sample dilution step. Several times the reaction media is cycled in and out of the SPR.

Measurements of IgG and IgM antibodies were conducted and interpreted according to the direction of the producer of VIDAS ToxoIgM and IgG kit: bioMerieux, Marcy- I'Etoil. France. When the assay is performed, the computer analyses the results automatically. For each sample examined, fluorescence is evaluated twice in the reagent strips reading cuvette. Prior to the introduction of SPR into the substrate, the initial reading is a background reading of the substrate cuvette. Following incubation of the substrate with the enzyme that remained on the interior of the SPR, the second reading is taken. By subtracting the background reading from the final result, the relative fluorescence value is determined by utilising the interpretation of lgM and lgG titers. Equivocal samples should be retested. If the interpretation remains equivocal, a new sample must be collected (Table 1).

VIDAS IgG Avidity

After initial detection of anti- Toxoplasma IgM and IgG, all samples were tested using the lgG avidity assay.

Table 1: Showing fluorescence site calculation for interpretation of IgG and IgM titer

Titer (IU/ml)	*Interpretation*
<4	Negative
4≤ titer <8	Equivocal
≥8	Positive

The assay includes a two-steps enzyme immunoassay sandwich approach as well as a final fluorescent detection step (ELFA). The SPR acts as both a solid phase and a pipetting device in the test. VIDAS IgG avidity uses a dual strip comprising one reference strip and one test strip. The sample to be tested, after dilution, is dispensed into both sample wells of the dual strips. The VIDAS Toxo-IgG Avidity Kit "bio-Merieux, Marcy-I'Etoile, France" was used to determine Toxoplasma IgG avidity according to the manufacturer's instructions and interpret the results. This test is also conducted by the fully automated VIDAS machine, which does the calculations and results in interpretation automatically. The index numbers given in Table 2 are used in the discrimination between the cases.

Table 2: The index numbers below are used in the discrimination between the cases

Avidity	*Interpretation*
Index < 0.200	Low avidity IgG
0.200 ≤ index < 0.300	Borderline avidity
index ≥0.300	Elevated avidity lgG

Results and Discussion

Comparative results of the VIDAS IgG.IgM and VIDAS IgG avidity tests are demonstrated in Tables 3 and 4.

From the seventy-eight samples subjected to these tests, nine were positive for Toxoplasma specific IgM antibodies and 33 for IeG antibodies, three of the nine (33.3%) IgM positive and four of the 33 (11.11%) IgG positive cases had low IgG avidity suggesting an active Toxoplasma infection. Also, two equivocal IgM (22.2%) and five equivocal IgG (75%) showed low avidity test results. They are also indicating an active infection, whereas four IgM positive (50%), twenty-four IgG positive (74.74%) and six equivocal IgM showed high avidity IgG antibodies, these indicating that the infection was acquired in the distant past.

Interestingly, two specimens from the 61 (3.27%) women with IgM negative showed low IgG avidity,

while 52 (85.24%) of the same group showed high IgG avidity. It is found that Spiramycin should be given to pregnant women who have a suspected acute toxoplasma infection.

Table 3: VIDAS IgM and VIDAS IgG avidity test results in pregnant women are compared

IgG Avidity	*Number of specimen with VIDAS IgM*		
	Positive n=9	*Equivocal n=8*	*Negative n=61*
Low	3(33.3%)	2(22.2%)	2(3.27%)
Borderline	2(15.7%)	0	7(11.47%)
High	4(50%)	6(787.8%)	52(85.24%)

Table 4: VIDAS IgG and VIDAS IgG avidity test results in pregnant women are compared

IgG Avidity	*Number of specimen with VIDAS IgG*		
	Positive n=33	*Equivocal n=6*	*Negative n=39*
Low	4(11.11%)	5 (75%)	Not done
Borderline	5(16.16%)	1(25%)	Not done
High	24(742.74%)	0	Not done

Detection of *T. gondii* during the early stage of pregnancy is very important to avoid intrauterine malformations. The common method used in the detection of Toxoplasma antibodies is through performing the ELISA test. This test is based on either the sero conversion of IgG or on the existence of positive Toxoplasma lgM antibodies (Agmas et al., 2015; Mwambe et al., 2013). The avidity-specific *T. gondii* IgG test can differentiate between the newly acquired and distant infection (Alvarado-Esquivel et al., 2011). The data from the present evaluation that has been carried out on 78 pregnant women indicated that the VIDAS IgG avidity test is an excellent method**.**

To differentiate between cases, where similar findings have been reported in the previous study (Andiappan et al., 2014), low avidity IgG (< 0.200) was determined only on three of nine and four of 33 pregnant women who had the lgM and IgG antibodies, respectively. They are also indicating an active infection, all of them led to acute Toxoplasma infection while the remaining other pregnant women with high avidity have chronic Toxoplasma infection.

The test of VIDAS IgG avidity was demonstrated to be useful in sera with ambiguous results with the VIDAS IgM and IgG tests. There may be ambiguities in the interpretation of some results, and there may be unclear interpretations, and this requires follow-up samples. Similar results have been indicated in a previous study (Alday and Doggett, 2017; Suzuki et al., 2001). With the avidity technique, it is possible to prove that women with Toxoplasma IgM at the time of pregnancy had not been infected recently. As a result, further IgG avidity measures are used to confirm or refute the IgM diagnosis (Cortina-Borja et al., 2010).

When IgG seroconversion or IgM positivity paired with low avidity of IgG is used to diagnose maternal primary infection, the mother should be referred to an obstetric consultation and amniotic fluid sampling for PCR, which is important for fetal infection verification. The avidity assay is placed behind the IgM and IgG tests in order to identify each patient using traditional infection markers. Furthermore, because the avidity test relies on the presence of particular IgG, the lack of IgG early after infection favours IgM testing in the first sample.

Conclusion

The assessment of the affinity of IgG in determining the time of infection, because these results indicate the examination of IgG affinity VIDAS type when used in combination with VIDAS assay for IgG, IgM antibodies to obtain confirmation of acute infection using a single serum sample of pregnant women**.**

References

Alday P.H. and J.S. Doggett (2017). Drugs in development for toxoplasmosis: Advances challenges and current status. *Drug Des Devel. Ther.*, **11:** 273-293.

Agmas, B., Tesfaye, R. and D.N. Koye (2015). Seroprevalence of *Toxoplasma gondii* infection and associated risk factors among pregnant women in Debre Tabor, Northwest Ethiopia. *BMC Research Notes*, **8(1):** 1-7.

Andiappan, H., Nissapatorn, V., Sawangjaroen, N., Chemoh, W., Lau, Y.L., Kumar, T. and V. Chandeying (2014). Toxoplasma infection in pregnant women: A current status in Songklanagarind hospital, southern Thailand. *Parasites & Vectors*, **7(1):** 1-7.

Alvarado-Esquivel, C., Estrada-Martínez, S. and O. Liesenfeld (2011). *Toxoplasma gondii* infection in workers occupationally exposed to unwashed raw fruits and vegetables: A case control seroprevalence study. *Parasites & Vectors*, **4(1):** 1-7.

Berredjem, H., Aouras, H., Benlaifa, M., Becheker, I. and M.R. Djebar (2017). Contribution of IgG avidity and PCR for the early diagnosis of toxoplasmosis in pregnant women from the North-Eastern region of Algeria. *African Health Sciences*, **17(3):** 647-656..

Cortina-Borja, M., Tan, H.K., Wallon, M., Paul, M., Prusa, A., Buffolano, W. and European Multicentre Study on Congenital Toxoplasmosis (EMSCOT) (2010). Prenatal treatment for serious neurological sequelae of congenital toxoplasmosis: An observational prospective cohort study. *PLoS Medicine*, **7(10):** e1000351.

Foroutan-Rad, M., Khademvatan S., Majidiani H., Aryamand, S., Rahim, F. and A.S. Malehi (2016). Seroprevalence of *Toxoplasma gondii* in the Iranian pregnant women: A systematic review and meta-analysis. *Acta Trop.*, **158:** 160-169.

Gutiérrez-Zufiaurre, N., Sánchez-Hernández, J., Muñoz, S., Marín, R., Delgado, N., Sáenz, M.C. and, J.A. García-Rodríguez (2004). Seroprevalence of antibodies against *Treponema pallidum, Toxoplasma gondii*, rubella virus, hepatitis B and C virus, and HIV in pregnant women. Enfermedadesinfecciosas y microbiologiaclinica, **22(9):** 512-516.

Kamani, J., Mani, A.U. and G.O. Egwu (2010). Seroprevalence of *Toxoplasma gondii* infection in domestic sheep and goats in Borno state, Nigeria. *Tropical Animal Health and Production*, **42:** 793-797.

Kompalic-Cristo, A., Frotta, C., Suárez-Mutis, M., Fernandes, O. and C. Britto (2007). Evaluation of a real-time PCR assay based on the repetitive B1 gene for the detection of *Toxoplasma gondii* in human peripheral blood. *Parasitology Research*, **101(3):** 619-625.

Mwambe, B., Mshana, S.E., Kidenya, B.R., Massinde, A.N., Mazigo, H.D., Michael, D. and U. Groß (2013). Seroprevalence and factors associated with Toxoplasma gondii infection among pregnant women attending antenatal care in Mwanza, Tanzania. *Parasites & Vectors*, **6(1):** 1-5.

Petersen, E., Pollak, A., and I. Reiter-Owona (2001). Recent trends in research on congenital toxoplasmosis. *International Journal for Parasitology*, **31(2):** 115-144.

Pomares, C. and J.G. Montoya (2016). Laboratory diagnosis of congenital toxoplasmosis. *Journal of Clinical Microbiology*, **54(10):** 2448-2454.

Rahbari, A.H., Keshavarz, H., Shojaee, S., Mohebali, M. and M. Rezaeian (2012). IgG avidity ELISA test for diagnosis of acute toxoplasmosis in humans. *The Korean Journal of Parasitology*, **50(2):** 99-102.

Ramzan, M., Akhtar, M., Muhammad, F., Hussain, I., Hiszczyńska-Sawicka, E., Haq, A.U. and M. AHafeez (2009). Seroprevalence of *Toxoplasma gondii* in sheep and goats in Rahim Yar Khan (Punjab), Pakistan. *Tropical Animal Health and Production*, **41(7):** 1225-1229.

Suzuki, L.A., Rocha, R.J. and C.L. Rossi (2001). Evaluation of serological markers for the immunodiagnosis of acute acquired toxoplasmosis. *Journal of Medical Microbiology*, **50(1):** 62-70.

Thiébaut, R., Leproust, S., Chêne, G. and R. Gilbert (2007). Effectiveness of prenatal treatment for congenital toxoplasmosis: A meta-analysis of individual patients' data. *The Lancet*, **369(9556):** 115-122.