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Association between *Toxoplasma gondii* Seropositivity and Memory Function in Non-Demented Older Adults

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Abstract

Toxoplasma gondii (*T. gondii*) seropositivity may be associated with decreased memory in older adults. To further investigate the association between *T. gondii* seropositivity and memory in non-demented older adults, we obtained serum samples from 114 non-demented older adults evaluated by the Alzheimer's Disease and Research Center at Washington University in St. Louis Missouri, USA. We determined *T. gondii* seropositivity and anti-*T. gondii* IgG antibody titer and examined associations with memory function while controlling for socioeconomic status, education level, age, and apolipoprotein E4 status. There were few associations between *T. gondii* seropositivity or anti-*T. gondii* IgG antibodies and memory, although there was some support suggesting an interaction between anti-*T. gondii* and sex. In the seropositive-only sample, there was an inverse relationship between anti-*T. gondii* titer and performance on the selective reminding test. Overall, we found little evidence of an association between impaired memory function and *T. gondii* seropositivity and anti-*T gondii* IgG antibodies in this sample of non-demented older adults.

Keywords

Toxoplasma gondii; toxoplasmosis; neurocognitive function; aging; memory

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1. Introduction

Multiple factors are associated with cognitive function and cognitive aging in older adults including sociodemographic variables, lifestyle, and diseases such as hypertension and diabetes (Gifford, et al., 2013, Stewart and Liolitsa, 1999, Zhang, et al., 2015). Furthermore, many of these factors may increase or protect against the risk of developing dementia (Xu, et al., 2015). In addition, the association between various infectious diseases, cognitive function, and risk for cognitive impairment and even dementia in older adults has been increasingly investigated (Barnes, et al., 2014, Katan, et al., 2013, Watson, et al., 2013).

Among the infectious diseases implicated in affecting cognition is *Toxoplasma gondii* (*T. gondii*), a protozoan parasite whose definitive host is any member of the cat family including domestic cats. Affecting approximately 12% of the United States' population (Jones, et al., 2014a), T. *gondii* can be transmitted to humans through the ingestion of oocysts (from cat feces) or through the ingestion of tissue cysts containing infectious bradyzoites from undercooked beef, pork, and other meats (Jones, et al., 2014b). Although *T. gondii* infection in humans often goes unnoticed in immunocompetent individuals, *T. gondii* tissue cysts can persist in human brain and muscle tissue (Montoya and Liesenfeld, 2004). *T. gondii* seropositivity increases with age, and 41% of older adults in the United States ages 60 to 90 years (median age = 69) may be infected (Mendy, et al., 2015). Further, *T. gondii* seropositivity appears to be associated with behavioral, cognitive, and neurological abnormalities in humans (Fekadu, et al., 2010, Flegr, 2015). Specifically, Kusbeci et al. (2011) found a significantly higher prevalence of *T. gondii* seropositivity in Alzheimer's disease compared to non-demented controls, suggesting *T. gondii* seropositivity may possibly contribute to dementia. However, a subsequent study by Perry et al. (2016) found no association between Alzheimer's disease and *T. gondii* seropositivity in a larger sample of older adults (114 control subjects and 105 Alzheimer's disease subjects). Thus, the role of *T. gondii* in the development of neurological diseases affecting memory remains unclear.

In one of the few studies investigating the association between *T. gondii* seropositivity and cognitive function in older adults, Mendy and colleagues (2015) analyzed a database from the Centers for Disease Control in the U.S. that included 4,485 participants with a median age of 69 years and compared the performance of subjects *T. gondii* seropositive and seronegative on two brief memory measures, one of which was the recall of three words from the Mini-Mental State Examination and the other the recall of a brief story. Controlling for covariates including age, sex, race-ethnicity, and education, they found that *T. gondii* seropositivity was associated with worsened immediate but not delayed recall. Gajewski et al. (2014) administered a much more detailed cognitive battery to a group of 42 elderly adults (mean age = 69.8 years) *T. gondii* seropositive and 42 *T. gondii* seronegative (mean age = 70.8) and found that verbal learning and delayed recall for a 15-word list were lower in the seropositive group.

Based on previously reported associations between *T. gondii* seropositivity and deficits in immediate memory, delayed recall, and verbal learning in older adults and the widespread distribution of *T. gondii* seropositivity in older adults, we sought to further investigate

associations between *T. gondii* seropositivity and memory function in a well characterized sample of older adults.

2. Methods

2.1 Study Sample

We obtained serum samples from 114 non-demented older adults evaluated by the Alzheimer's Disease and Research Center (ADRC) at Washington University in St. Louis Missouri, USA, one of many Alzheimer's Disease Centers across the United States funded by the National Institute of Aging and the same sample we used for a control group in a previous study (Perry, et al., 2016). With few exceptions, all subjects had clinical dementia rating (CDR) (Morris, 1993) scores of 0 (no dementia) and the cognitive status of each subject was not suggestive of dementia (McKhann, et al., 2011). Accordingly, all subjects in the current study had been defined as non-demented control subjects.

2.2 Determination of T. gondii Seropositivity

To identify *T. gondii* seropositivity, we assayed serum samples by quantitative ELISA for the presence of IgG anti-*T. gondii* antibodies using a commercially available ELISA kit (GenWay, BioTech). We did all assays according to the manufacturer's recommended protocol and ran all samples in duplicate. In accordance with the manufacturer's guidelines, we considered samples to be seropositive for *T. gondii* when the *T. gondii* IgG-class antibody titer was greater than 35 IU/mL. We considered samples below 30 IU/mL to be seronegative according to the manufacturer's guidelines. We assayed samples in the equivocal range, measuring between 30 and 35 IU/mL, a second time. Positive controls included those provided by the kit manufacturer as well as an additional patient sample that was confirmed seropositive through the use of a kit produced by a different manufacturer (Abcam). In addition to positive controls, we assayed three to four patient samples from each plate on a subsequent plate to confirm reproducibility of the results. We diluted any samples measuring outside of the standard curve (generated on each plate) five and ten-fold and then assayed them again with results adjusted according to the dilution factor. The researcher conducting all assays was blind to all demographic variables. The same researcher performed all ELISA assays in an effort to reduce any potential user variation.

The presence of IgG anti-*T gondii* antibodies in the serum in the absence of IgM antibodies is indicative of a past infection with *T gondii*, whereas the presence of IgM antibodies is considered indicative of an acute or reactivated *T. gondii* infection. In an effort to distinguish between acute and a past infection with *T. gondii*, we assayed all samples seropositive or borderline positive for IgG antibodies to determine the titer of IgM anti-*T. gondii* antibodies using a commercially available IgM specific ELISA (GenWay, BioTech) following the protocol provided by the manufacturer. According to the protocol, we obtained the *Toxoplasma* IgM index by dividing the mean values of each sample by the calibrator mean value considering a *Toxoplasma* IgM index greater than 1 as positive for IgM antibodies to *T. gondii*, an index between 0.91 and 0.99 as equivocal, and an index below 0.90 as negative. As with the IgG assays, we ran all IgM assays in duplicate, and the researcher performing these assays was blind to all demographic data. We ran a positive and negative control in

each experiment, and all results were within the index range for positive and negative controls.

2.3 Cognitive Function

Available measures of memory included the Logical Memory Story A (LM IA), Immediate and Delayed recall (LM IIA) from the Wechsler Memory Scale-Revised (Wechsler, 1987), Associate Learning from the Wechsler Memory Scale (Wechsler, 1973) and the selective reminding test (SRT) (Grober, et al., 1988). We included the total recall score from the SRT, which includes the number of items recalled on the three cued recall trials. All of these tests measure explicit memory.

2.4 Estimate of Socioeconomic Status

In this sample, the Hollingshead two-factor index of social position provided an estimate of socioeconomic status, with "1" on the scale representing higher socioeconomic status with the range of possible values on the scale being from 1 to 5 (Hollingshead and Redlich, 1958). For the analyses, we dichotomized this scale to high and low SES for ease of interpretation. Specifically, we considered scores from 1–3 as high and scores 4 and 5 as low.

2.5 Statistical Analyses

We used descriptive statistics to characterize the sample according to age, sex, socioeconomic status, educational attainment, apolipoprotein E4 (ApoE) status, average antibody titer, and performance on each of the memory tasks. We used a natural-logarithm transformation to normalize the distribution of the antibody titer and *t*-tests to compare scores on each measure between the *T. gondii* seropositive and seronegative groups. In the statistical analysis, we used two methods: linear regression and a post-hoc Bayesian analysis, both modeled without an intercept (Rencher and Schaalje, 2008). To ensure the assumptions of non-intercept regression were not violated, we centered both the age and antibody titers around zero.

We first used linear regressions to predict scores in each memory task using two models. The first model included the dichotomous measure of *T. gondii* seropositivity or seronegativity, and the second included the log-transformed, continuous measure of anti-*T. gondii* IgG antibody titers. Both models included sex, socioeconomic status, education level, age, and apolipoprotein E4 (ApoE) status (presence or absence of E4 allele) as control variables. We included ApoE status as a control variable due to its known association both with Alzheimer's disease and cognitive decline (Dorey, et al., 2014). Specifically, Levy et al. (2004) found those carrying an E4 allele of ApoE had lower scores on Wechsler Memory Scale – Revised Logical Memory II. Given these findings, we wanted to control for the potential effect ApoE status may have on memory function and thus included it in our analyses. In each model, the differences in sample size are due to missing data on the dependent variables.

In addition, we analyzed the associations between the *T. gondii* seropositivity and performances on the memory tasks using the same models as mentioned previously but with

an interaction included as we were interested in the effect of titer and sex. Also, in post-hoc analyses, we used the same linear regressions and Bayesian models to predict scores in each memory task using anti-*T. gondii* IgG antibody titers for *T. gondii* seropositive samples separately.

Because our sample size was relatively small, we performed a post-hoc power analysis for both the main effects models and the *T. gondii* seropositivity and sex interaction models with alpha set at .05 using G*power 3.1 software (University of Düsseldorf, http://www.gpower.hhu.de).

Because of the potential for alpha inflation due to multiple comparisons in linear regression, we also used a Bayesian analysis to further evaluate our findings. In Bayesian analyses, there is not a problem with alpha inflation from multiple comparisons because the parameters have a joint posterior distribution, and Bayesian analyses are constructed on understanding the limits of applicability instead of hypothesis testing (Gelman, et al., 2013).

In order to predict scores in each memory task, we fit a Bayesian generalized linear model using two sets of regressors, as we did in the above linear regression. The first set included the dichotomous measure of latent toxoplasmosis (Tx), and the second included the log-transformed, continuous measure of anti-*T. gondii* IgG antibody titers (LnTx). Both models included sex, socioeconomic status, education level, age, and ApoE status (presence or absence of E4 allele) as control variables. As we assumed the missing data was missing at random, the differences in sample size are due to taking out the missing variables.

Because each memory task is in a bounded region, we transformed each score y in the Bayesian analysis to be $y^*=\frac{y}{b}$ where b is the maximum score given the memory task. When the extreme scores (either a 0 or the maximum) was reached, the transformation for all scores was then $y^*=(\frac{y}{b}(n-1)+0.5)/n$ as suggested by Smithson and Verkuilen (2006). After the transformation, we have

$$y \sim Beta(\mu, \phi)$$

where μ is the mean and ϕ is the precision parameter. Additionally, for the generalized linear model, we used the linear predictor $\eta = X\boldsymbol{\beta}$ where $\boldsymbol{\beta}$ are the coefficients for the independent and control variables. Finally we used logit link function $g(\cdot)$;thus $\mu=g^{-1}(X\boldsymbol{\beta})=\frac{e^{X\boldsymbol{\beta}}}{1+e^{X\boldsymbol{\beta}}}$

To finish specifying the Bayesian model, we put a normal prior on the $\boldsymbol{\beta}$ coefficients and an inverse gamma prior on the precision parameter ϕ. In particular, we had

$$\beta_j \sim N(0, 5)$$
$$\phi \sim IG(3, 20)$$

We then used Metropolis-Hastings algorithm to sample from the joint posterior (Gelman, et al., 2013). Both trace plots and the stationarity test based on Heidelberger and Welch (1983) indicated convergence of the simulation.

To compare the models fit with either the dichotomous or continuous measure of *T. gondii* seropositivity as well as the differing link functions, we used the Deviance Information Criterion (DIC) metric, which is specifically useful for hierarchical Bayesian models (Spiegelhalter, et al., 2002). We first calculated the deviance of each posterior sample,

$$D(\mu,\phi) = -2\log\left(\frac{\Gamma(\phi)}{\Gamma(\mu\phi)\Gamma((1-\mu)\phi)} y^{\mu\phi-1}(1-y)^{(1-\mu)\phi-1}\right)$$

The average of all the deviance scores is denoted $\overline{D}$. Then the overall DIC is

$$DIC = 2\overline{D} - D(\overline{\mu},\overline{\phi})$$

where $\overline{\mu}$ and $\overline{\phi}$ are the mean of the posterior samples.

3. Results

Of the 114 subjects in this study, 38 (33.3%) were *T. gondii* seropositive. The average age of the seropositive group was 81.1 years, compared to an average of 79.0 years in the seronegative group, and all subjects included in the analysis were white. The two groups did not differ significantly by age ($p = 0.1$), sex ($p = 0.5$), education ($p = 0.4$), socioeconomic status ($p = 0.3$), or percent with at least one ApoE E4 allele ($p = 1.0$). As expected, the anti-*T. gondii* IgG concentration was significantly higher in the *T. gondii* seropositive group compared to the seronegative group ($p < 0.001$) (Table 1). Only one subject was seropositive for anti-*T. gondii* IgM-class antibodies, whom we removed from the analysis. The prevalence of hypertension, diabetes, heart disease, thyroid disease, alcohol abuse, tobacco use, stroke, and hypercholesterolemia did not significantly differ between the seropositive and seronegative groups.

WMS Logical Memory IA Immediate, WMS Logical Memory IIA Delayed, WMS Associate Learning, and the Total score from the SRT, uncorrected for any control variables, did not differ in the overall sample between subjects *T. gondii* seropositive and seronegative (Table 1). Additionally, scores uncorrected for any control variables on measures of memory did not differ significantly between males and females (Table 2).

3.1 Linear Regression Results

In the linear-regression analyses, T. *gondii* seropositivity was not associated with any of the memory measures in the overall sample in the model including control variables. However, statistical power ranged from 45 to 87 percent (Table 3) suggesting that the probability of false negative results of the tests was high.

In the seropositive-only interaction analyses, anti-*T. gondii* IgG titer was not associated with any of the memory measures for seropositive samples, nor was the titer-sex interaction (Table 4). Again, the power of the tests was very low (22% to 78%).

However, there was a significant interaction between anti-*T. gondii* antibody titer and sex such that the slope of anti-*T-gondii* antibody titer for males was more positive than the slope for females (i.e., on Logical Memory IA and Logical Memory IIA in the overall sample (Table 5). However, presence or absence of *T. gondii* seropositivity did not predict any memory test scores in the overall sample. There was low power for the SRT model. However, the power in the other models ranged from 55.1 to 84.8 percent.

3.2 Bayesian analysis results

The analyses presented in Table 5 were also estimated as Bayesian models. The DIC scores for the four outcomes by anti-*T. gondii* antibody titer and *T. gondii* seropositivity are presented in Supplemental Table 1. For the Logical Memory tests (both immediate (IA) and delayed recall (IIA)) and for the SRT, the model including anti-*T. gondii* antibody titer has a slightly better fit. On the other hand, the Associate Learning test had a better model fit for *T. gondii* seropositivity (Supplemental Table 1).

Parameters from the Bayesian analysis and their corresponding posterior density 95% credible interval are listed in the Supplemental Table 2. In particular, only the SRT test had an interaction between sex and anti-*T. gondii* antibody titer. For the SRT, higher titers in males was associated with higher scores with an odds ratio of 1.22, meaning there is a 22% increase in one unit of the scores for every one unit increase of titers of toxoplasmosis for males. Females, on the other hand, only had a 2% increase. That is, males had more of an improvement with higher titers. Note, this was when the entire sample (seropositive and seronegative) was included.

For the seropositive-only sample in the Bayesian analysis, the antibody titer for the SRT task had an odds ratio of .38, meaning there was a 62% decrease in one unit of the memory score for every one unit increase in titers of toxoplasmosis of those participants that were seropositive (Supplemental Table 3). However, there was no significant titer-sex interaction in the Bayesian seropositive only sample.

4. Discussion

In this well characterized sample of 114 older adults without dementia, there were few associations between memory function and *T. gondii* seropositivity in both linear regression and Bayesian models. Both seropositive and seronegative groups were well matched on sociodemographic and medical variables. In the linear regression models without interaction terms, there were no associations between either *T. gondii* seropositivity or anti-*T. gondii* antibody titer levels and any of the memory tasks. In the entire sample, there was a significant interaction, however, between anti-*T. gondii* antibody titer and sex, indicating either that males with higher anti-*T. gondii* IgG antibody titers had better performance than did males with lower titers or that males with lower titers had worse performance on the Logical Memory IA and IIA tasks. We did not find this association in the model looking at

presence or absence of toxoplasmosis (i.e. seropositivity), nor was there an interaction between antibody titer (i.e. serointensity) and sex in the linear-regression models in the seropositive-sample only. Further, the results of the Bayesian models showed no main effects for *T. gondii* seropositivity or antibody titer on any of the memory tasks, although there was an interaction between sex and *T. gondii* antibody titer on the SRT task, with higher titers of anti-*T. gondii* antibodies in males associated with higher SRT scores. In the Bayesian model for the seropositive-only sample, higher antibody titer was associated with lower scores on the SRT task, suggesting that higher serointensity may be associated with worse performance on the SRT. However, we did not find any association in the Bayesian analyses in the seropositive-sample only and the other three memory tasks.

Although we had expected that *T. gondii* seropositivity would be negatively associated with memory function, our finding of limited support suggesting that *T. gondii* seropositivity is associated with better memory function on some of the memory tasks in males is consistent with a study of young adults where performance on a task of executive function modulated by dopamine was positively associated with *T. gondii* seropositivity (Stock, et al., 2014). In this regard, the SRT we used in our study has a strong executive component not present in the other three memory tests we used. However, we found different results in the regression models, where in males only increased anti-*T. gondii* antibody titers were associated with higher performance on the immediate (Logical Memory IA) and delayed recall (Logical Memory IIA) portions of the Logical Memory subtest of the Wechsler Memory Scale rather than on the SRT. These findings may have different interpretations, depending on whether higher anti-*T. gondii* antibody titer represents greater serointensity or whether lower titers of anti-*T. gondii* in seropositive subjects indicates a greater interval since the initial infection. If higher titers indicate higher serointensity, our findings may suggest limited evidence (i.e. on two of four memory measures) of an association between *T. gondii* and better memory function in males. If lower titers that are still in the range considered positive for the presence of latent toxoplasmosis reflect longer time since infection, there would be limited evidence of worse memory function in low anti *T. gondii* IgG males, that is, in males infected long before the onset of the present study. However, in the Bayesian analysis in the seropositive-only sample containing both females and males, there was an inverse relationship between antibody titer and performance on the SRT with higher titers associated with worse performance.

In addition to IgG seropositivity (i.e. presence of latent infection), some studies suggest higher anti-*T. gondii* titers are often associated with worse cognitive performance (Mendy, et al., 2015). Similarly, behavioral or psychiatric symptoms associated with chronic *T. gondii* infection are typically correlated with higher rather than lower titers (Groer, et al., 2011, Okusaga, et al., 2011, Pedersen, et al., 2012). Moreover, serointensity but not seropositivity differentiated between patients with schizophrenia and healthy controls (Emelia, et al., 2012) and also between patients with depression with or without a history of suicide attempts (Arling, et al., 2009). However, even though many studies suggest seropositivity for latent toxoplasmosis is associated with worse outcome, whether it be cognitive or psychiatric, other studies suggest that seropositivity may not be purely detrimental. For example, Flegr et al. (2012) analyzed males and females separately and found that while seropositive females did more poorly on one of several cognitive measures compared to seronegative females,

seropositive males did equally well on most cognitive measures and actually did better than seronegative males on two of the cognitive measures. Similarly, Stock et al. (2014) found that seropositivity for *T. gondii* was associated with better performance on a measure of action control. Finally, there is another possibility to be considered. Havlicek et al. (2001) found that seropositive subjects with lower rather than higher titers were the ones that performed differently from seronegative controls and they suggested that lower titers in seropositive subjects reflect longer duration of latent *T. gondii* infection, an interpretation broadly consistent with a study by Konishi (1989), who demonstrated that longitudinally those with initially higher IgG antibody levels tended to demonstrate decreases in IgG antibody levels over time. Still, many of the subjects demonstrated no significant change over a few years, and some subjects with initially lower titer levels showed increased titers with time. Further, an anamnestic decrease in anti-*T. gondii* IgG titers could have led to false negatives in our study of older adults resulting in misclassification of *T. gondii* infection status, which could be a reason for our finding few if any differences in memory function associated with *T. gondii* seropositivity in older adults. Older subjects infected well before the study could have normal anti-*T. gondii* due to the anamnestic antibody response (Kodym P, et al., 2001). However, since our present data are cross-sectional, we cannot directly address chronicity of infection, and we cannot assume that seropositive subjects with lower titers are those with the longest duration of latent toxoplasma infection. Furthermore, we have no way to determine whether any of the subjects had either been exposed to the infection more than once or whether an existing infection had been reactivated at some point. Recent animal studies have demonstrated that situational factors such as stressors can reactivate a chronic *T. gondii* infection with conversion of *T. gondii* bradyzoites to tachyzoites (Shen, et al., 2016). In animals, an increase in tachyzoites in chronic *T. gondii* infection is associated with increases in IgG and IgA but not IgM antibodies (Singh, et al., 2010).

Possible reasons for the differences between our findings and those of Mendy et al. (2015) and Gajewski et al. (2014), who found a negative impact of toxoplasmosis on memory function in older adults, could be due to age differences between studies in that our sample had a mean age of 81.1 and 79.0 years for the seropositive and seronegative groups, respectively, compared to the median age in the study by Mendy et al. (2015) of 69 years and in the Gajewski et al. (2014) study a mean age of 70.8 and 69.8 years in the seropositive and seronegative groups, respectively. Because our sample was on average approximately ten years older, there could have been a higher death rate in the seropositive group compared to the seronegative group resulting in a loss of those subjects with potential memory deficits associated with *T. gondii* seropositivity. In this regard, we note that *T. gondii* seropositivity has been negatively associated with health status, particularly related to immune and digestive disorders (Flegr and Escudero, 2016, Flegr, et al., 2014) and has been associated with morbidity and mortality (Lykins, et al., 2016). Also, because we excluded participants with a diagnosis of dementia, the key feature of which is often memory impairment, we may have had a sample biased towards average memory performance. In addition, both Mendy et al. (2015) and Gajewski et al. (2014) used different tests to evaluate memory, which may have evaluated different aspects of memory function than did the tests of memory function we used. Still, the tests used in our study are well known and often used clinically for

diagnostic purposes in the evaluation of age-related memory decline. Finally, the sample we used had relatively high levels of education, 15.3 years and 14.8 years of education in the seropositive and seronegative groups, respectively, which may have protected against any negative effects of *T. gondii* seropositivity on memory (Gale, et al., 2015).

How *T. gondii* seropositivity might account for both improved and worsened memory function in older adults is unclear. A possible mechanism may relate to the increased production of dopamine associated with *T. gondii* seropositivity, as dopamine has been associated with better working and verbal memory function (Henry and Sherwin, 2012). Specifically, Prandovszky et al. (2011) showed higher levels of dopamine in cyst-containing neural cells in mice infected with Toxoplasmosis, and McConkey et al. (2013) proposed that the parasite's ability to increase dopamine also affects catecholaminergic neurons and the behaviors associated with them. Another study analyzing performance on a cognitive task modulated by the dopamine system found that *T. gondii* seropositivity in a group of 25-year olds was associated with better function (Stock, et al., 2014).

Although the subjects in this study were well characterized and were assessed with standard measures of memory function, several limitations in addition to those already discussed require consideration in interpreting these findings. Low statistical power may have led to false negatives in the analyses. In addition, the study design was cross-sectional, and we did not have information about the time of initial infection with *T. gondii*. Similarly, we did not have information about the length of time subjects had been *T. gondii* seropositive.

5. Conclusion

In conclusion, in this sample of 114 older female and male subjects, taking into account the limitations associated with this study including low statistical power, there was little evidence of an association between *T. gondii* seropositivity and anti-*T. gondii* IgG antibody titer and memory function. However, there was some evidence of an interaction between *T. gondii* and memory performance in males and also evidence that higher anti-*T. gondii* titers in the seropositive-only sample was associated with worse performance on the SRT.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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6. Appendix

Table 1

Characteristics of the toxoplasmosis positive and negative groups.

Characteristics	Positive	Negative	Total	*t* [CI], p-value**
Sample Size	38	76	114	
Age (years), mean, SD, [range]	81.1, 6.5, [70.4–94.3]	79.0, 7.6, [62.1–98.3]	79.7, 7.3, [62.1–98.3]	−1.5 [−5.0, 0.7], 0.1
Women (%)	45	51	49	−0.7 [−0.1, 0.3], 0.5
Education (years), mean, SD	15.3, 2.9	14.8, 3.0	15.0, 3.0	−0.9 [−1.7, 0.6], 0.4
SES (Hollingshead Index), mean, SD	3.9, 1.0	3.8, 1.0	3.8, 1.0	−1.0 [−0.6, 0.2], 0.3
Prevalence of E4 allele (ApoE) (%)	26	27	27	0.0 [−0.2, 0.2], 1.0
Anti-*T. gondii* IgG Ab concentration IU/mL, mean, SD, [range]	293.5, 244.7, [35.2–940.8]	13.2, 7.0, [0.3–28.9]	106.6, 193.0, [0.3–940.8]	−10.0 [−335.7, −224.8], <0.001
MMSE total score, mean, SD	28.5, 1.7	28.7, 1.4	28.7, 1.5	0.8 [−0.3, 0.8], 0.4
WMS Logical Memory IA Immediate, mean, SD	14.4, 4.0	13.9, 4.6	14.1, 4.4	−0.4 [−2.5, 1.6], 0.7
WMS Logical Memory IIA, mean, SD	13.5, 4.5	12.9, 5.0	13.1, 4.8	−0.5 [−2.8, 1.7], 0.6
WMS Associate Learning, mean, SD	14.6, 3.0	14.4, 3.9	14.5, 3.6	−0.3 [−1.6, 1.2], 0.8
Free and Cued Selective Reminding Test				
Total score, mean, SD	47.8, 0.5	47.7, 0.8	47.7, 0.7	−0.3 [−0.4, 0.3], 0.7

*No significant differences between groups for hypertension, diabetes, heart disease, thyroid disease, alcohol consumption, tobacco use, stroke, hypercholesterolemia.

**T-tests were performed between the groups which were seropositive or seronegative for *T. gondii.*

Table 2

Memory test scores (M, SD) for males and females.

Memory measure	Males	Females	Total	*t* [CI], p-value**
WMS Logical Memory IA, Immediate, mean, SD	13.45, 0.7	14.7, 0.6	14.1, 1.0	−1.3 [−3.2, 0.7], 0.2
WMS Logical Memory IIA, mean, SD	12.3, 0.8	13.9, 0.7	13.1, 0.5	−1.5 [−3.7, 0.6], 0.1
WMS Associate Learning, mean, SD	13.8, 0.5	15.2, 0.5	14.5, 0.7	0.1 [−2.7, 0.0], 0.1

Memory measure	Males	Females	Total	*t* [CI], p-value**
Free and Cued Selective Reminding Test				
Total score, mean, SD	47.7, 0.1	47.7, 0.1	47.7, 1.0	0.0 [−0.3, 0.3], 1.0

** T-tests were performed between the male and female groups

Table 3

Main effects models of anti-*T. gondii* antibody titer or presence of *T. gondii* seropositivity and each memory test score for the whole group: Unstandardized coefficients (standard errors) from linear regression.

	Logical Memory IA		Logical Memory IIA		Associate Memory		Selective Reminding Test	
	b	SE	b	SE	b	SE	b	SE
Antibody titer	.030	(.02)	.033	(.02)	.000	(.02)	−.005	(.02)
Male	−.088*	(.04)	−.100*	(.05)	−.064	(.04)	−.015	(.04)
SES (Hollingshead Index)								
High	.070	(.05)	.066	(.05)	.091*	(.04)	.093*	(.04)
Education								
More than High School	.580***	(.03)	.542***	(.04)	.687***	(.03)	.951***	(.03)
Age	−.002	(.02)	.005	(.02)	−.005	(.02)	.009	(.02)
ApoE (presence of E4)	.032	(.05)	.049	(.05)	.029	(.05)	.092*	(.04)
R^2	.13		.16		.16		.08	
N	81		81		107		91	
Power	.627		.735		.871		.453	
Presence of Latent Toxoplasmosis	.021	(.04)	.027	(.05)	.014	(.04)	.010	(.04)
Male	−.082	(.04)	−.094*	(.05)	−.065	(.04)	−.016	(.04)
SES (Hollingshead Index)								
High	.070	(.05)	.067	(.05)	.091*	(.04)	.093*	(.04)
Education								
More than High School	.574***	(.04)	.534***	(.04)	.682***	(.04)	.947***	(.03)
Age	.000	(.00)	.001	(.00)	−.001	(.00)	.001	(.00)
ApoE (presence of E4)	.019	(.05)	.034	(.05)	.029	(.05)	.094*	(.04)
R^2	.11		.13		.16		.08	
N	81		81		107		91	
Power	.543		.627		.871		.453	

Abbreviations: SE = Standard error.

* $p < .05$,

** $p < .01$,

*** $p < .001$.

Table 4

Interaction effects (titer-sex interaction) models of anti-*T gondii* antibody titer and each memory test score for seropositive samples only: Unstandardized coefficients (standard errors) from linear regression.

	Logical Memory IA		Logical Memory IIA		Associate Memory		Selective Reminding Test	
	b	**SE**	**b**	**SE**	**b**	**SE**	**b**	**SE**
Antibody titer	−.083	(.12)	.076	(.10)	−.040	(.06)	−.111	(.10)
Male	−.045	(.07)	−.036	(.07)	−.061	(.04)	−.068	(.05)
SES (Hollingshead Index)								
High	.118	(.08)	.134	(.09)	.201***	(.04)	195***	(.05)
Education								
More than High School	.204**	(.02)	.188***	(.15)	.252***	(.01)	.353***	(.01)
Age	−.004	(.01)	−.006	(.05)	−.008*	(.00)	.001	(.00)
ApoE (presence of E4)	.058	(.09)	−.058	(.10)	−.102	(.05)	−.037	(.07)
Titer−sex interaction	.126	(.14)	.154	(.15)	.012	(.08)	−0.42	(.11)
R^2	.19		.17		.47		.32	
N	28		28		37		31	
Power	.245		.221		.781		.470	

Abbreviations: SE = Standard error.

* $p < .05$,

** $p < .01$,

*** $p < .001$.

Table 5

Interaction models (titer-sex, Toxoplasmosis) of the relationship between anti-*T. gondii* antibody titer or presence of *T. gondii* seropositivity and each memory test score for whole group: Unstandardized coefficients (standard errors) from linear regression.

	Logical Memory IA		Logical Memory IIA		Associate Memory		Selective Reminding Test	
	b	**SE**	**b**	**SE**	**b**	**SE**	**b**	**SE**
Antibody titer	−.010	(.03)	−.012	(.03)	−.020	(.03)	−.030	(.03)
Male	−.091*	(.04)	−.103*	(.04)	−.066	(.04)	−.017	(.04)
SES (Hollingshead Index)								
High	.083	(.04)	.081	(.05)	.095*	(.04)	.102*	(.04)
Education								
More than High School	.573***	(.03)	.534***	(.03)	.684***	(.03)	945***	(.03)
Age	.000	(.02)	.008	(.02)	−.004	(.02)	.008	(.02)
ApoE (presence of E4)	.044	(.05)	.062	(.05)	.032	(.05)	.099*	(.04)
Titer-sex interaction	.081*	(.04)	.094*	(.04)	.040	(.04)	.053	(.04)
R^2	.15		.18		.16		.08	
N	81		81		107		91	
Power	.667		.762		.848		.422	

	Logical Memory IA		Logical Memory IIA		Associate Memory		Selective Reminding Test	
	b	SE	b	SE	b	SE	b	SE
Presence of Latent Toxoplasmosis	−.043	(.06)	−.052	(.07)	−.014	(.06)	−.021	(.06)
Male	−.130*	(.05)	−.153*	(.06)	−.085	(.05)	−.037	(.05)
SES (Hollingshead Index)								
High	.083	(.05)	.083	(.05)	.096*	(.05)	.098*	(.04)
Education								
More than High School	.593***	(.04)	.557***	(.04)	.689***	(.04)	.955***	(.03)
Age	.000	(.00)	.001	(.00)	−.001	(.00)	.001	(.00)
ApoE (presence of E4)	.027	(.05)	.044	(.05)	.031	(.05)	.096*	(.04)
Toxoplasmosis-sex interaction	.128	(.09)	.157	(.10)	.054	(.09)	.059	(.08)
R^2	.1		.15		.16		.08	
N	81		81		107		91	
Power	.551		.667		.848		.422	

Abbreviations: SE = Standard error.

*$p < .05$,

**$p < .01$,

***$p < .001$.

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Highlights

- Latent Toxoplasmosis has been associated with decreased memory in some studies.
- In this sample, there were few associations between memory and latent toxoplasmosis.
- In seropositive participants, higher anti-*T. gondii* titers were inversely associated with memory.