



Neurocognitive effects of latent *Toxoplasma gondii* infection in immunocompromised people with HIV

Research Article


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Abstract

Toxoplasma gondii (*T. gondii*) is a neurotropic parasite that establishes latent infection in the central nervous system (CNS), and may alter behaviour and contribute to neuronal dysfunction. However, its impact on cognitive performance and CNS pathophysiological alterations in people with HIV (PWH) remains unclear. A cross-sectional study of adult PWH was conducted, assessing latent *T. gondii* infection through serum IgG levels in the absence of neurotoxoplasmosis. Neurocognitive impairments were assessed through neurocognitive testing across 6 domains, along with depressive and anxiety symptoms evaluation, and educational attainment. CNS pathophysiological alterations were assessed through amyloid- β 1-42, total tau, phosphorylated tau, neopterin, and S-100 β quantification in cerebrospinal fluid (CSF). Fifty-eight PWH were included, and *T. gondii* seropositivity was detected in 46.5% of participants (27/58). Overall, cognitive performance was largely comparable between groups, although subtle, non-significant declines were observed across several domains. *T. gondii*-seropositive individuals demonstrated a faster completion of Trail Making Test Part B ($\beta = -35.79$ sec; 95% CI: -67.78 to -3.86), lower educational attainment ($\beta = -1.92$ years; 95% CI: -3.76 to -0.09), without different levels of CSF biomarkers for neuronal-synaptic degeneration, Alzheimer's pathology, beta-amyloid deposition, macrophage-derived inflammation and glial activation-degeneration. In PWH with low CD4 counts, latent *T. gondii* infection was not associated with overt cognitive impairment or detectable CNS pathophysiological alterations. Instead, an atypical profile emerged, combining faster task-switching with lower educational attainment and subtle, non-significant declines in other domains. These findings highlight the complex nature of *T. gondii*-host interactions and need for longitudinal studies to clarify long-term neurocognitive outcomes.

Introduction

Toxoplasma gondii (*T. gondii*) is an obligate intracellular parasite with marked neurotropism, infecting up to one-third of the global population (Montoya and Liesenfeld, 2004). Its ability to establish chronic infections in immune-privileged sites like the central nervous system (CNS) enhances its persistence. In the CNS, the parasite switch to a low-replicative form, encysting within neurons and glial cells. Tissue cysts are impermeable to drugs and impervious to host immune responses, typically resulting in a silent, lifelong infection.

A growing body of research highlights the multifaceted impact of *T. gondii* infection on the nervous system, spanning molecular, cellular and behavioural alterations in both *in vitro* and *in vivo* studies. *In vitro* studies indicate that *T. gondii* affects the γ -aminobutyric acid system (MacRae et al., 2012). In animal models, *T. gondii* has been shown to affect the glutamate systems (David et al., 2016), to induce phosphorylation of tau protein and apoptosis of hippocampal neurons (Tao et al., 2020), and to induce marked behavioural changes. In particular, *T. gondii* appears to specifically alter certain behavioural patterns, such as reducing the predator aversion through epigenetic modulation in the amygdala, thereby facilitating its transmission to felid hosts (Vyas et al., 2007; Hari Dass and Vyas, 2014; Poirotte et al., 2016). Therefore, motor performance, exploration of novel environments, host risk-taking tendencies or even suicidal tendencies can be altered (Vyas et al., 2007; Hari Dass and Vyas, 2014; Poirotte et al., 2016).

Interestingly, although infection in immunocompetent humans is typically asymptomatic and they are no longer part of a predator-prey trophic cycle with felids, accumulating evidence

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strongly associate *T. gondii* with neurodegenerative and psychiatric disorders. Specifically, *T. gondii* has been associated with self-directed violence in psychiatric subjects, schizophrenia, aggression and impulsivity, suicide attempts and traffic accidents, as well as Alzheimer's disease (Torrey et al., 2012; Cook et al., 2015; Coccaro et al., 2016; Nayeri Chegeni et al., 2019; Sutterland et al., 2019). Furthermore, *T. gondii* has been found in children and older individuals with cognitive impairment (Gajewski et al., 2014; Mendy et al., 2015).

In people with HIV (PWH), where low cognitive performance are well-documented and known to impair quality of life (Heaton et al., 2010; Saylor et al., 2016; Trunfio et al., 2018), *T. gondii* is one of the most prevalent opportunistic infection of the CNS (Wang et al., 2017), potentially fueling chronic immune activation, indirect neuronal damage and, possibly, neurocognitive deficits (Escobar-Guevara et al., 2023; Diaz et al., 2024; Manuel et al., 2025).

Therefore, the present study aims to evaluate the effect of latent *T. gondii* infection on neurocognitive performances and CNS pathophysiological alterations in a group of PWH without neurotoxoplasmosis.

Materials and methods

Study population

A descriptive, single-centre, cross-sectional study involving adult PWH was conducted. The enrolled subjects were admitted to the Infectious Diseases Department of Amedeo di Savoia Hospital, Turin, Italy, for neurocognitive evaluation and for cerebrospinal fluid (CSF) and serum specimens' collection. The samples were appropriately stored at -80°C until further use. In this cohort, both the neurocognitive assessment and the biological specimens' retrieval were performed as part of routine clinical care for neurological symptoms. In the present study, the latent *T. gondii* infection was defined as the presence of anti-*T. gondii* IgG in the absence of overt clinical symptoms. The definition of latent *T. gondii* infection aligns with current literature, which, despite progress in elucidating the mechanisms of latency and developing novel latency-specific biomarkers, continues to rely on these criteria (Egorov et al., 2021; Licon et al., 2023; Diaz et al., 2024; Getzmann et al., 2024; Laguardia et al., 2024; Silva et al., 2024; Montazeri et al., 2025; Robert et al., 2025). Additionally, individuals with a confirmed diagnosis of neurotoxoplasmosis, as defined by the presence of radiological findings on Magnetic Resonance Imaging compatible with the disease, the presence of *T. gondii* IgG, and no evidence of alternative aetiologies (e.g. lymphoma, cryptococcoma or abscesses), were excluded from the study. HIV-associated neurocognitive disorders (i.e. HAND) were assessed using the previously described Frascati criteria, categorizing HAND according to the extent of impairment in everyday functioning as asymptomatic neurocognitive impairment (ANI), mild neurocognitive disorder (MND) or HIV-associated dementia (HAD) (Antinori et al., 2007). The Medical Ethical Committee of the University of Turin approved each phase of the study, and written informed consent was obtained from all study participants.

Anti-*T. gondii* IgG antibodies

Anti-*T. gondii* IgG levels were evaluated with chemiluminescent immunoassay (CLIA) (LIAISON[®], DiaSorin, Saluggia, Italy) on

participants' serum samples. According to manufacturer's instruction anti-*T. gondii* IgG concentrations were categorized as follows: negative (below 7.2 IU mL^{-1}), dubious (between 7.2 and 8.8 IU mL^{-1}) and positive (above or equal to 8.8 IU mL^{-1}).

Assessment of neurocognitive impairments

Neurocognitive performance was evaluated through full neurocognitive testing (NCT). NCT was carried out by a trained neuropsychologist and included 12 neurocognitive tests for 6 cognitive domains: Memory (Corsi Test, Serial Repetition Disyllabic Words Test), Attention/working memory (Trail Making Test part B, Stroop Colour Test Time, Stroop Colour Interference Error), Executive functions (Frontal Assessment Battery, Verbal fluency test), Ideomotor speed (Digit symbol, Trail Making Test Part A), Visuospatial Capabilities (Rey-Osterrieth Complex Figure Copy) and Motor Functioning (Grooved Pegboard Test Dominant Hand, and Grooved Pegboard Test Non-Dominant Hand) (Antinori et al., 2007; Trunfio et al., 2018). Depressive and anxiety symptoms were assessed using the Beck Depression Inventory II (BDI-II) and Hamilton Anxiety Rating Scale (HAM-A). Educational attainment, expressed as cumulative years of school attendance, was recorded as an indicator of cognitive reserve, which has recently been associated with reduced risk of cognitive impairments (Zhong et al., 2024).

Assessment of central nervous system involvement

CSF biomarkers were used for assessing neuronal-synaptic degeneration (total tau, i.e. T-tau) (Soares et al., 2025), Alzheimer's pathology (phosphorylated tau, i.e. P-tau) (Ashton et al., 2022), beta-amyloid deposition (amyloid- β 1-42) (Theodorou et al., 2025), macrophage-derived inflammation (neopterin) (Miyae et al., 2024), as well as glial activation and degeneration (S-100 β) (Papuć and Rejdak, 2020). Briefly, amyloid- β 1-42, T-tau and P-tau were quantified immunoenzymatically (Fujirebio diagnostics, Malvern, PA, USA); neopterin and S-100 β through validated ELISA assays (DRG Diagnostics kit Marburg, Germany and DIAMETRA Srl Spello, Italy, respectively). Reference values were T-tau $< 300\text{ pg mL}^{-1}$ (in patients aged 21–50), $< 450\text{ pg mL}^{-1}$ (in patients aged 51–70) or $< 500\text{ pg mL}^{-1}$ (in older patients); P-tau $< 61\text{ pg mL}^{-1}$; amyloid- β 1-42 $> 500\text{ pg mL}^{-1}$; neopterin $< 1.5\text{ ng mL}^{-1}$; S-100 β $< 380\text{ pg mL}^{-1}$.

Statistics

Eligible individuals were categorized in two groups based on the detectability of anti-*T. gondii* IgG, and continuous variables were compared using a Mann-Whitney U test, whereas categorical variables were compared using a Fisher's exact. A generalized linear model was used to assess the effect of *T. gondii* infection on the neurocognitive impairments and CSF biomarkers of CNS pathophysiological alterations correcting for CD4 T cell counts nadir, CSF HIV viral load, potential neurotoxic ARTs, and months from HIV diagnosis. A *P*-value of less than 0.05 was considered statistically significant, all statistical analyses were performed using SPSS version 30.0.0, and results were graphed with GraphPad Prism 10.3.1.

Results

Study population and anti-*T. gondii* IgG prevalence

A total of 58 adult PWH were enrolled in the present study (Table 1). The median age of the cohort was 48.4 years (IQR 42.7–54.1), with the majority being male (75.9%) and of Caucasian ethnicity (93.1%). At the time of enrolment, 56.9% of participants ($n = 33/58$) were receiving combination antiretroviral therapy (i.e. cART). Among PWH receiving cART, the majority was receiving triple therapy NRTI + PI (48.5%) or NRTI + INSTI (18.2%). Importantly, most individuals of the cohort had a CD4 + T cell count below 500 cells mm^{-3} , with a median of 120 cells mm^{-3} (IQR 39–434), and consequent low CD4/CD8 ratio (median 0.2; IQR 0.1–0.7). The median plasma HIV RNA level was 3 \log_{10} (copies mL^{-1}) (IQR 1.3–5.5), while the median CSF HIV RNA level was 2.1 \log_{10} (copies mL^{-1}) (IQR 1.3–3.6). The median time since HIV diagnosis was 22.3 months (IQR: 1.4–151.6), and the majority of subjects either presented an ANI (65.5%) or did not have any HAND (25.9%). None of the participants included in the study had a history of toxoplasmic encephalitis (TE)/cerebral toxoplasmosis (CTX).

Next, *T. gondii* co-infection was evaluated by quantifying anti-*T. gondii* IgG antibodies in serum samples, which revealed a prevalence of 46.5% ($n = 27/58$) within our cohort. Participants were subsequently stratified into two groups based on the detectability of anti-*T. gondii* IgG in those without any detectable IgG levels (i.e. Tg-IgG –, $n = 31$) and those with any detectable IgG levels (i.e. Tg-IgG+, $n = 27$). Among Tg-IgG+ individuals, the median anti-*T. gondii* IgG titer was 175 IU mL^{-1} (28–400 IQR). The two groups were comparable in terms of age, sex, ethnicity, and comorbidities (Table 1). No statistically significant differences were observed between Tg-IgG+ and Tg-IgG– individuals with respect to CD4 + T cell counts, CD4/CD8 ratios, cART regimens, cART with neurotoxic potential, months from diagnosis, nor HAND. Similarly, HIV RNA levels in both plasma and CSF specimens did not differ significantly between the two groups (Table 1).

T. Gondii and neurocognitive impairments in PWH

Full NCT is shown in Figure 1 (panels A–F). The most involved domain appeared to be the visuospatial capabilities as measured by the Rey-Osterrieth Complex Figure Copy test. In the unadjusted model, *T. gondii* seropositivity was associated with poorer visuospatial performance (estimate: –6.90; 95% CI: –12.95 to –0.86), and, after adjusting for the relevant covariates, a comparable negative trend, though not statistically significant, persisted (estimate: –5.42; 95% CI: –11.79 to 0.95; P -value = 0.095) (Figure 1D). Within the attention/working memory domain, despite comparable performance on measures of inhibitory control and selective attention (i.e. Stroop Colour Inference Error, P -value = 0.826, and Stroop Colour Test Time, P -value = 0.707), subjects with co-infection presented a faster completion of the Trail Making Test Part B (estimate: –35.79 seconds; 95% CI: –67.78 to –3.86; $P = 0.028$) (Figure 1F). Second, depressive and anxiety symptoms were assessed through BDI-II and HAM-A scores; however, having the co-infection did not associate with heightened anxiety or depression (Figure 1G). Third, educational attainment was assessed as an indicator of cognitive reserve, and individuals with co-infection showed a significant lower education attainment (estimate: –1.92; 95% CI: –3.76 to –0.09; P -value = 0.040) (Figure 1H). This atypical pattern may reflect a subtle but widespread negative impact of co-infection on neurocognitive performance.

T. Gondii and CSF biomarkers of CNS pathophysiology in PWH

To investigate potential biological correlates of neuronal-synaptic degeneration, Alzheimer's pathology, beta-amyloid deposition, macrophage-derived inflammation, as well as glial activation and degeneration associated with *T. gondii* seropositivity, five established CSF biomarkers were quantified: amyloid- β 1-42, T-tau, P-tau, neopterin and S-100 β . In this cohort, the median values of CSF amyloid- β 1-42 (968 pg mL^{-1} [IQR: 730.5–1122]), T-tau (152 pg mL^{-1} [IQR: 88.3–242.9]), P-tau (35 pg mL^{-1} [IQR: 24.7–44]), neopterin (1.1 ng mL^{-1} [IQR: 0.8–3.5]) and S100 β (105.1 pg mL^{-1} [64.2–165.5]) were all within the normal range. Indeed, reference values were as follow: i.e. T-tau < 300 pg mL^{-1} (in patients aged 21–50), <450 pg mL^{-1} (in patients aged 51–70), or <500 pg mL^{-1} (in older patients); P-tau < 61 pg mL^{-1} ; amyloid- β 1-42 > 500 pg mL^{-1} ; neopterin < 1.5 ng mL^{-1} ; S-100 β < 380 pg mL^{-1} . Furthermore, neither in the unadjusted nor in the adjusted model was *T. gondii* seropositivity associated with differences in CSF biomarker levels (Figure 1I). Specifically, the group with co-infection was associated with the following estimates: amyloid- β 1-42 (estimate: 64.86; 95% CI: –78.75 to 208.48; P -value = 0.376), T-tau (estimate: –14.94; 95% CI: –81.94 to 52.06; P -value = 0.662), P-tau (estimate: 0.22; 95% CI: –8.59 to 9.02; P -value = 0.961), Neopterin (estimate: –1.28; 95% CI: –4.89 to 2.34; P -value = 0.488), S-100 β (estimate: 57.59; 95% CI: –24.55 to 139.74; P -value = 0.169).

These findings suggest that latent *T. gondii* infection is not associated with detectable alterations in the levels of CSF selected biomarkers.

Discussion

T. gondii is a neurotropic parasite with a marked affinity for the CNS. In individuals with impaired immune functions, it can cause neurotoxoplasmosis, a life-threatening condition with severe neurological manifestations. When the infection is effectively controlled, patients may remain seropositive without overt neurological symptoms. Although latent *T. gondii* infection has long been considered largely harmless, three decades of research now indicate otherwise, revealing a subtle but widespread impact on cognitive performance, behaviour and population-level morbidity (Latifi and Flegr, 2025). This issue is particularly relevant in PWH, who not only exhibit a higher prevalence of *T. gondii* infection but also are at risk of cognitive impairment despite antiretroviral therapy (Heaton et al., 2010; Wang et al., 2020). However, data addressing the impact of latent *T. gondii* infection on neurocognitive outcomes in PWH remain limited. Therefore, in an attempt to address this knowledge gap, this study aimed to investigate the impact of latent *T. gondii* infection on neurocognitive performance and CSF biomarkers of CNS pathophysiology in PWH with low CD4 T cell counts without neurotoxoplasmosis.

Interestingly, the global seroprevalence of *T. gondii* is estimated at 31% (95% CI: 28–34), with substantial heterogeneity across regions (e.g. 42% in Africa and 25% in Asia) (Sengupta et al., 2025). Prevalence rates are higher among immunocompromised subjects, with a recent pooled estimate of 42% (95% CI: 34–49) (Rahmanian et al., 2020). In the context of HIV infection, the global prevalence of *T. gondii* co-infection in PWH has been estimated at 35.8% (95%: 30.8–40.7), with the highest rates observed in sub-Saharan Africa and low-income countries (Wang et al., 2017). Consistently, our findings revealed a seroprevalence of *T. gondii* infection of 46.5% among PWH with low CD4 T cell counts, aligning with previous

Table 1. Study population

	Overall (n = 58)	Tg-IgG + (n = 27)	Tg-IgG - (n = 31)	P-value* (Tg-IgG + vs Tg-IgG -)
Age, years (median [IQR])	48.4 (42.7–54.1)	51 (44.7–59.1)	46.7 (42.4–52)	0.185
Sex (n [%])				0.378
Male	44 (75.9)	22 (81.5)	22 (71.0)	
Female	14 (24.1)	5 (18.5)	9 (29.9)	
Ethnicity (n [%])				0.785
Caucasian	54 (93.1)	25 (92.6)	29 (93.5)	
Southamerican	3 (5.2)	1 (3.7)	2 (6.4)	
African	1 (1.7)	1 (3.7)	0 (0)	
CD4 T cells count (cells mm ⁻³)	120 (39–434)	291 (19–463)	107 (56.7–307)	0.631
Co-morbidities (n [%])				
Hypertension	4 (6.9)	0 (0)	4 (12.9)	0.053
Ischemic heart disease	1 (1.7)	0 (0)	1 (3.2)	0.346
Non-ischemic heart disease	1 (1.7)	0 (0)	1 (3.2)	0.346
Chronic kidney disease	2 (3.4)	2 (7.4)	0 (0)	0.123
Liver disease	7 (12.1)	5 (18.5)	2 (6.4)	0.159
Neurological disease	8 (13.8)	4 (14.8)	4 (12.9)	0.833
Gastrointestinal disease	10 (17.2)	5 (18.5)	5 (16.1)	0.810
Diabetes	2 (3.4)	1 (3.7)	1 (3.2)	0.921
Lung disease	18 (31.0)	7 (25.9)	11 (35.5)	0.433
Cancer	5 (8.6)	2 (7.4)	3 (9.7)	0.759
Ratio CD4/CD8 (median [IQR])	0.2 (0.1–0.7)	0.3 (0–0.7)	0.2 (0.1–0.4)	0.578
Plasma HIV RNA, log ₁₀ (cp mL ⁻¹), (median [IQR])	3 (1.3–5.5)	1.7 (1.3–5.2)	5 (1.8–5.6)	0.135
CSF HIV RNA, log ₁₀ (cp mL ⁻¹), (median [IQR])	2.1 (1.3–3.6)	1.3 (1.3–3.8)	2.2 (1.4–3.6)	0.234
Any cART (n [%])	33 (56.9)	18 (66.7)	15 (48.4)	0.297
cART (n [%])				0.485
2 NRTI + 1 INSTI	6/33 (18.2)	2 (7.4)	4 (12.9)	
2 NRTI + 1 PI	16/33 (48.5)	10 (37)	6 (19.3)	
NNRTI	1/33 (3.2)	0 (0)	1 (3.2)	
NRTI + PI + INSTI	2/33 (6.1)	2 (7.4)	0 (0)	
NRTI + NNRTI	1/33 (3.2)	0 (0)	1 (3.2)	
PI + INSTI	3/33 (9.1)	2 (7.4)	1 (3.2)	
PI + NRTI	1/33 (3.2)	1 (3.7)	0 (0)	
OTHERS	3/33 (9.1)	1 (3.7)	2 (6.4)	
Anti- <i>T. gondii</i> IgG, IU mL ⁻¹ , (median [IQR])	-	175 (28–400)	-	-
Months from HIV diagnosis, (median [IQR])	22.3 (1.4–151.6)	35 (3–187.3)	15.8 (0.8–138.5)	0.351
HAND (n [%])				0.627
None	15 (25.9)	8 (29.6)	7 (22.6)	
ANI	38 (65.5)	16 (59.3)	22 (71)	
MND	4 (6.9)	2 (7.4)	2 (6.4)	
HAD	1 (1.7)	1 (3.7)	0 (0)	

IQR, interquartile range; CSF, cerebrospinal fluid; cART, combination antiretroviral therapy; NRTI, nucleoside reverse transcriptase inhibitors; INSTI, integrase strand transfer inhibitors; PI, protease inhibitors; NNRTI, non-nucleoside reverse transcriptase inhibitors; HAND, HIV-associated neurocognitive disorders; ANI, HIV-associated asymptomatic neurocognitive impairment; MND, HIV-associated mild neurocognitive disorder; HAD, HIV-associated Dementia. *Statistical analyses: Mann–Whitney U test or Fisher exact test, as appropriate; P-value < 0.05 considered statistically significant.

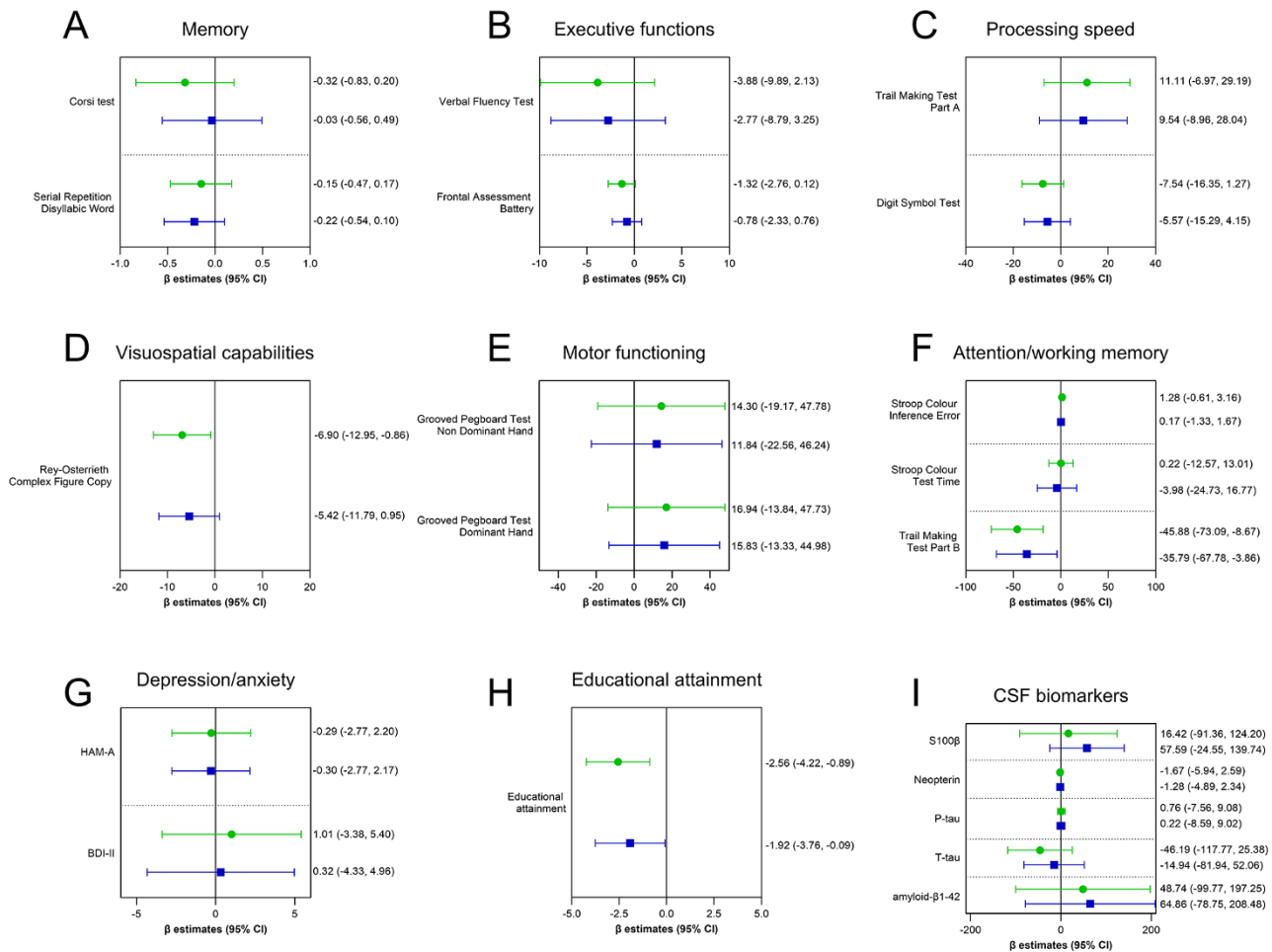


Figure 1. Effect of *T. Gondii* infection on neurocognitive performance and CFS markers of CNS pathophysiological alterations. Lines represent the estimated β coefficients (mean differences between IgG+ and IgG-) with corresponding 95% confidence intervals (CI), derived from the generalized linear model. The green-coloured line with circle represents the unadjusted β coefficients, whereas the blue-coloured line with square represents the adjusted β coefficients for the relevant covariates (CD4 T cell count nadir, CSF HIV viral load, potential neurotoxic ARTs, months from HIV diagnosis). Educational attainment was additionally corrected for age. P-value < 0.05 is considered statistically significant.

reports associating immunosuppression as well as HIV infection with higher *T. gondii* prevalence.

Furthermore, although the prevalence of HAND has decreased with cART (Mastrorosa et al., 2023), neurocognitive impairment remains common, with an estimated overall prevalence of 42.6% (ANI 23.5%, MND 13.3%, HAD 5%) (Wang et al., 2020). However, the potential impact of HAND on our study measures may be considered negligible. Indeed, on one hand, the majority of individuals fell within the spectrum of either no HAND or ANI. On the other hand, emerging evidence indicates that conventional neurocognitive assessments may overestimate disease burden, suggesting that the high reported prevalence of HAND may not fully reflect clinically meaningful impairment (Nightingale et al., 2023): while TE may be crucial for legacy symptoms, the role of *T. gondii* infection in active HIV-associated brain injury is unknown. Accurate diagnosis in PWH therefore requires simultaneous consideration of cognitive symptoms, low performance on cognitive tests, and abnormalities on neurological investigations (Nightingale et al., 2023), and we did not detect CSF biomarkers alterations.

In our cohort, *T. gondii* infection did not appear to strikingly affect the neurocognitive performance, except for a faster completion of the Trail Making Test Part B and a reduced educational

attainment. Importantly, a faster performance on the Trail Making Test Part B, which occurred in the absence of better outcomes in the Stroop Colour tests, should be interpreted with caution, and argue against a generalized improvement in executive control. Indeed, the complex and multifactorial mechanisms underlying the performance on this test have already been shown (Chan et al., 2015; Varjadic et al., 2018; Recker et al., 2022). Furthermore, *T. gondii* has been associated self-directed violence in psychiatric populations, including schizophrenia, aggression and impulsivity, suicide attempts and traffic accidents (Torrey et al., 2012; Cook et al., 2015; Coccaro et al., 2016; Sutterland et al., 2019). Therefore, our findings may reflect differences in response strategies, impulsivity or altered risk-taking behaviour rather than cognitive benefits. The trends observed across the other cognitive domains are consistent with subtle, non-significant but widespread decline in cognitive performance, as observed in the visuospatial performance. The lack of significant alterations in CSF biomarkers of neuronal-synaptic degeneration, Alzheimer's pathology, beta-amyloid deposition, macrophage-derived inflammation or glial activation and degeneration align with these cognitive findings. Moreover, these biological data are further supported by previous *in vitro* and *in vivo* studies, which have shown more subtle biological mechanisms

associated with *T. gondii* infection, such as modulation of the γ -aminobutyric acid and glutamate systems, or epigenetic changes in the amygdala (MacRae et al., 2012; Hari Dass and Vyas, 2014; David et al., 2016). Therefore, the faster completion of the Trail Making Test Part B and the reduced educational attainment appear to occur in the absence of detectable CNS pathophysiological alterations.

The atypical pattern of *T. gondii*–host interaction observed in our cohort is not surprising. Indeed, whereas *T. gondii* infection is often associated with adverse cognitive outcomes, emerging evidence suggests that under specific conditions it may exert neuroprotective or compensatory effects on the brain. For example, in a mouse model of cerebral ischemia, chronic *T. gondii* infection was linked to reduced brain damage and improved neurobehavioral performance, potentially through the upregulation of hypoxia-inducible factor 1- α (HIF-1 α) and vascular endothelial growth factor (VEGF), both of which may enhance cerebral resilience (Lee et al., 2020). Therefore, the mechanisms underlying *T. gondii*–human interaction may be more complex than initially thought. Additionally, recent findings have shown that PWH with latent toxoplasmic infection, as defined by the presence of anti-*T. gondii* IgG, do not exhibit different cognitive performance as compared to PWH without latent toxoplasmic infection, as defined by the absence of anti-*T. gondii* IgG (Diaz et al., 2024). Notably, cognitive impairment was significantly worse among PWH diagnosed with toxoplasmic encephalitis. Our study confirms and extends these observations in a more heterogeneous cohort. While we did not find statistically significant associations between *T. gondii* infection and cognitive impairment, the observed trajectories suggest a subtle but widespread trend towards poorer performance across most cognitive domains. These patterns may be linked to the significantly lower educational attainment observed in this group. Indeed, previous research has identified education as a key predictor of cognitive impairment (Zhong et al., 2024). Furthermore, longitudinal data show that cognitive performance in PWH, regardless of whether they had latent *T. gondii* infection or encephalitis, remained stable over a 7-year follow-up, despite the highest prevalence of cognitive impairments in the domains of verbal, executive function, learning, recall, working memory, processing speed and motor functions in those with encephalitis (Diaz et al., 2024). However, despite our results align with the literature showing subtle but widespread effects of latent *T. gondii* infection on cognitive performance and behaviour (Latifi and Flegr, 2025), the present study cannot exclude the occurrence of increased cognitive impairments over a longer follow-up.

Several limitations must be acknowledged when interpreting the results of this study. First, the relatively small sample size as well as the heterogeneity of the study population may have limited the statistical power. These constraints are especially relevant in the context of complex cognitive phenotypes, which are influenced by numerous biological, clinical and environmental factors. Moreover, the cross-sectional design of the study precludes any causal inferences about the directionality or temporality of the observed associations. Longitudinal data would be needed to determine whether *T. gondii* seropositivity predicts cognitive decline or modulates neurodegenerative progression over time. Furthermore, the mechanisms underlying the absence of overt clinical symptoms in our cohort cannot be determined; e.g. this could reflect *T. gondii* infection before HIV acquisition or the effect of trimethoprim/sulfamethoxazole prophylaxis. Lastly, the findings of the present study should be interpreted within the context of a predominantly male

PWH population with low CD4 T cell counts, but future studies will be valuable to clarify potential sex-specific differences (Latifi and Flegr, 2025), as well as the impact of higher CD4 T cell counts.

In summary, these initial findings in PWH with low CD4 counts and a likely *T. gondii* latent infection, suggest that they do not exhibit markedly worse cognitive performance, although subtle declines are suggested across domains. These individuals show lower educational attainment and a faster completion of the Trail Making Test Part B without evidence of detectable CNS pathophysiological alterations. These findings highlight the complex, context-dependent effects of *T. gondii* on the brain. Given the study's limitations, particularly the sample size and cross-sectional design, caution is warranted. Nonetheless, the results call for further research into how chronic infections interact with cognition in vulnerable populations. In this context, there is a growing effort to identify circulating plasma biomarkers that could facilitate large-scale studies and enable less invasive monitoring of CNS pathophysiological alterations (Prins et al., 2022; Vrillon et al., 2024), as well as to better define latent *T. gondii* infection (Egorov et al., 2021; Licon et al., 2023; Robert et al., 2025). Longitudinal, mechanistic studies with larger cohorts are needed to clarify whether *T. gondii* contributes to cognitive outcomes in HIV, and under what conditions such effects may be harmful, neutral, or adaptive.

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Artificial intelligence. ChatGPT5 was used to proofread the main text.

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