Latent toxoplasmosis is associated with neurocognitive impairment in a cohort of young adults with chronic HIV infection from Romania

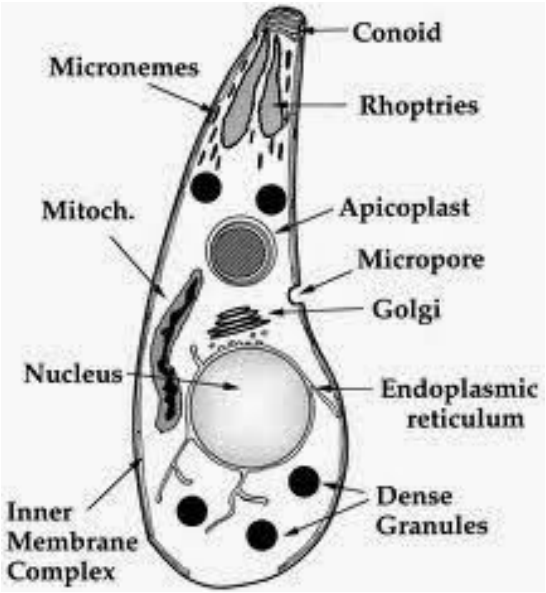
Authors: Ene L[1], Marcotte T[2] Grancea C[3], Temereanca A[3], Luca A[1], Bharti A[2], Umlauf A[2], Achim CL[2], Letendre S [2], Ruta SM[3]

[1] "Dr. Victor Babes" Hospital for Infectious and Tropical Diseases, Bucharest, Romania

[2] HIV Neurobehavioral Research Program, University of California San Diego, San Diego, USA

[3] " Stefan S. Nicolau" Institute of Virology, Bucharest, Romania

Parasites and us – who is in control?



"Toxoplasma is a kind of marvel pathogen. It can infect everything which is warm blooded and
it can be as silent as non existent in a system".

?

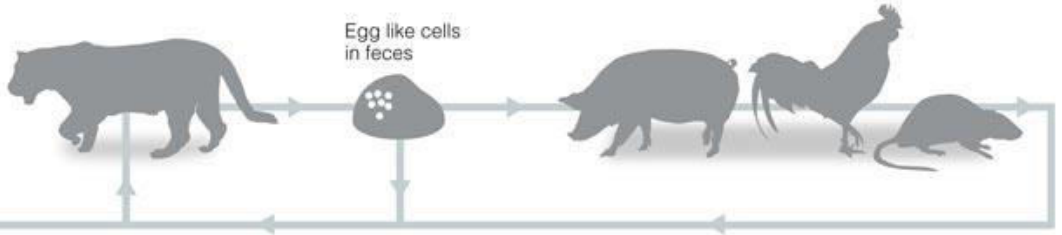
Michael W. Black, and John C. Boothroyd Microbiol. Mol. Biol. Rev. 2000;64:607-623

A Ubiquitous Pathogen That Keeps Its Host Healthy

The single-celled pathogen Toxoplasma gondii can enter the most protected parts of its host body while remaining largely undetected. In most cases it lives as a harmless tenant, but in fetuses or in people with compromised immune systems it can cause severe damage.

Host to host

While Toxoplasma can infect humans, other mammals, and birds, its relationship with cats is unique. Only in cats can the pathogen reproduce sexually to create egg like cells.

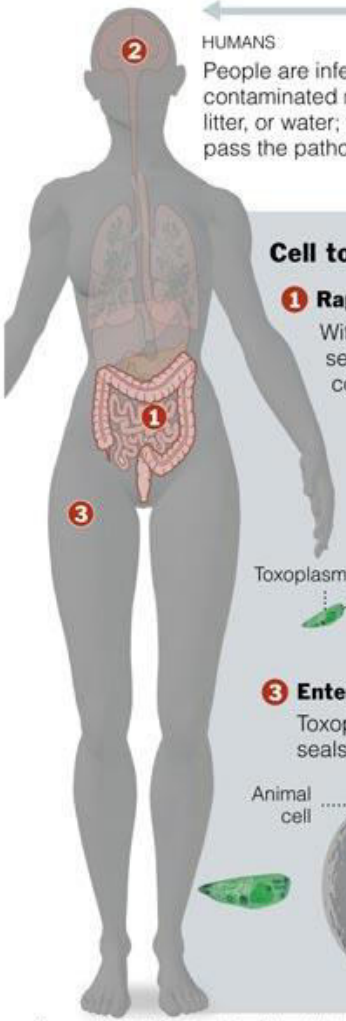


HUMANS
People are infected from contaminated meat, soil, kitty litter, or water; women can pass the pathogen to a fetus.

PRIMARY HOST: CATS
Toxoplasma can mature and sexually reproduce in members of all cat species.

EGG LIKE CELLS (OOCYSTS)
100 million oocysts can be shed in the droppings of a cat after a single infection. They can survive in the soil for more than a year.

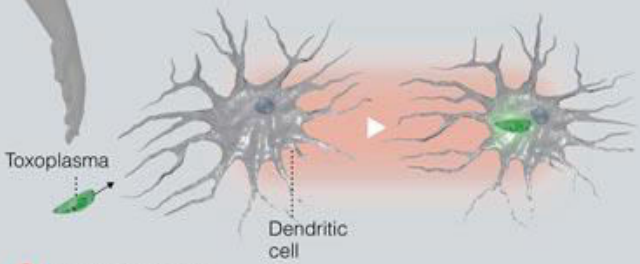
OTHER HOSTS
Animals that ingest Toxoplasma can develop cysts in their tissues. The cysts remain infective if their meat is not cooked.



Cell to cell

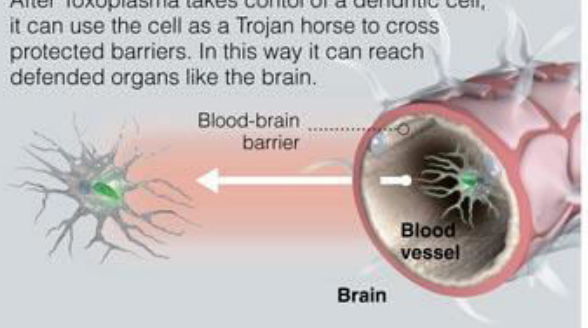
1 Rapid spread

Within hours of infection, Toxoplasma can move to widely separated parts of the body. It does this by entering and controlling dendritic immune cells in the intestine.



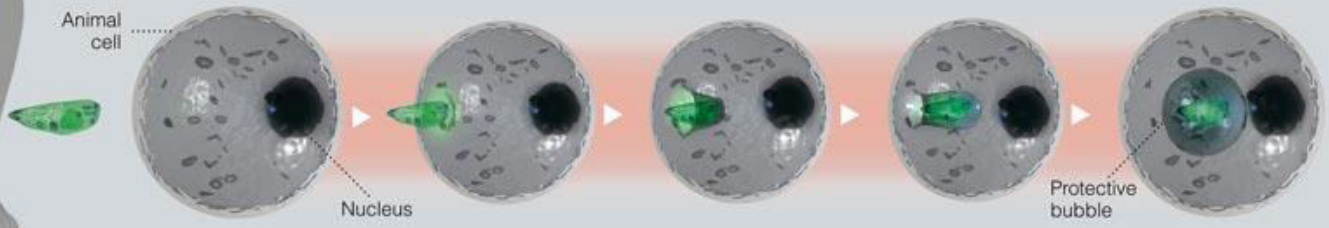
2 Crossing protected barriers

After Toxoplasma takes contol of a dendritic cell, it can use the cell as a Trojan horse to cross protected barriers. In this way it can reach defended organs like the brain.



3 Entering a cell

Toxoplasma can infect almost every type of cell. It enters by pushing against the membrane and pulling it over itself. The cell seals behind, leaving the pathogen in a protective bubble.

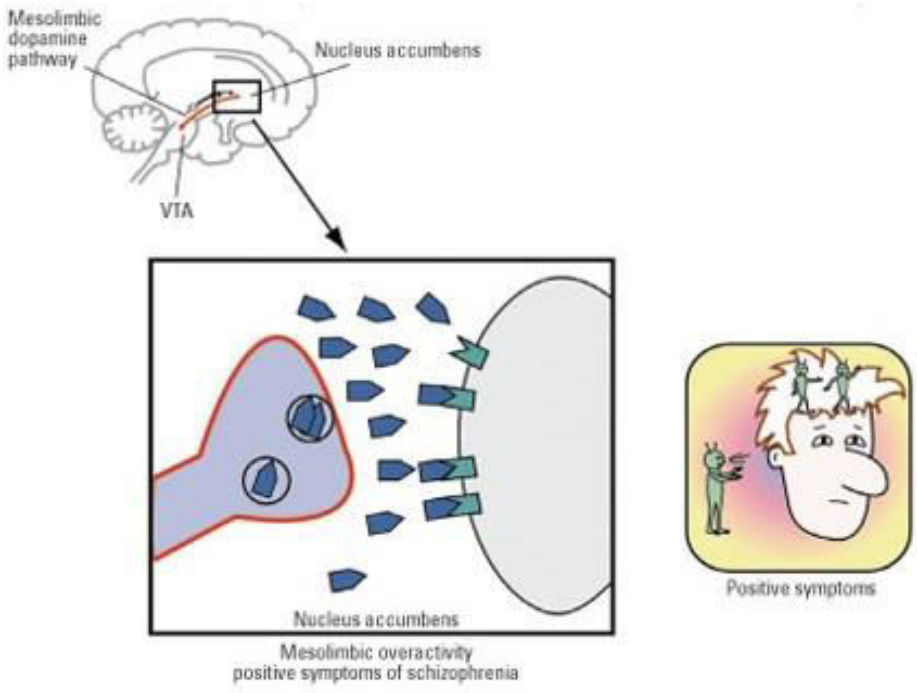


Sources: Antonio Barragan, Karolinska Institute and Swedish Institute for Infectious Disease Control; J. P. Dubey, Journal of the American Veterinary Medical Association

Graham Roberts/The New York Times

- High quantities of dopamine released by *T.gondii* may be responsible for clinical behavioural changes (*PLoS One* 2011,**6**:e23866)
- The presence of dopamine induces increased production of tachyzoites and destruction of the cysts walls *J Parasitol* 2012,**98**:1296-1299)
- Toxoplasma up regulated the miR-132 and is associated with changes in dopamine receptor signalling (Neuroscience. 2014 May 30;268:128-38)
- During its life cycle, Toxoplasma interacts with about 3000 genes and proteins, including susceptibility genes for Alzheimer disease, Schizophrenia, and mood disorders (J Pathog. 2013, 965046)

The mesolimbic dopamine hypothesis of positive symptoms of schizophrenia



VTA=ventral tegmental area.

Stahl SM. *CNS Spectr.* Vol 12, No 4. 2007.

Latent Toxoplasma infection was linked to psychiatric conditions & behavioral changes

- Schizophrenia Yolken, R. H., et al. (2009). Parasite Immunol 31(11): 706-715.
- Bipolar disorder Dickerson, F., et al. (2014" Bipolar Disord 16(2): 129-136; Hamdani, N., et al. (2013). J Affect Disord 148(2-3): 444-448
- Major depression, generalised anxiety disorder, panic disorder Gale, S. D., et al. (2014). Folia Parasitol (Praha) **61**(4): 285-292
- The additional diagnosis of a personality disorder in psychiatric patients Hinze-Selch, D., et al. (2010). Folia Parasitol (Praha) 57(2): 129-135
- Trait aggression and impulsivity Cook, T. B., et al. (2015). J Psychiatr Res 60: 87-94.
- Increased risk of traffic accidents Yereli, K., et al. (2006) Forensic Sci Int 163(1-2): 34-37; Flegr, J., et al. (2009) BMC Infect Dis 9: 72;
- Reccurent headache in adolescents Prandota, J., et al. (2014). "Recurrent headaches may be caused by cerebral toxoplasmosis." World J Clin Pediatr 3(3): 59-68.

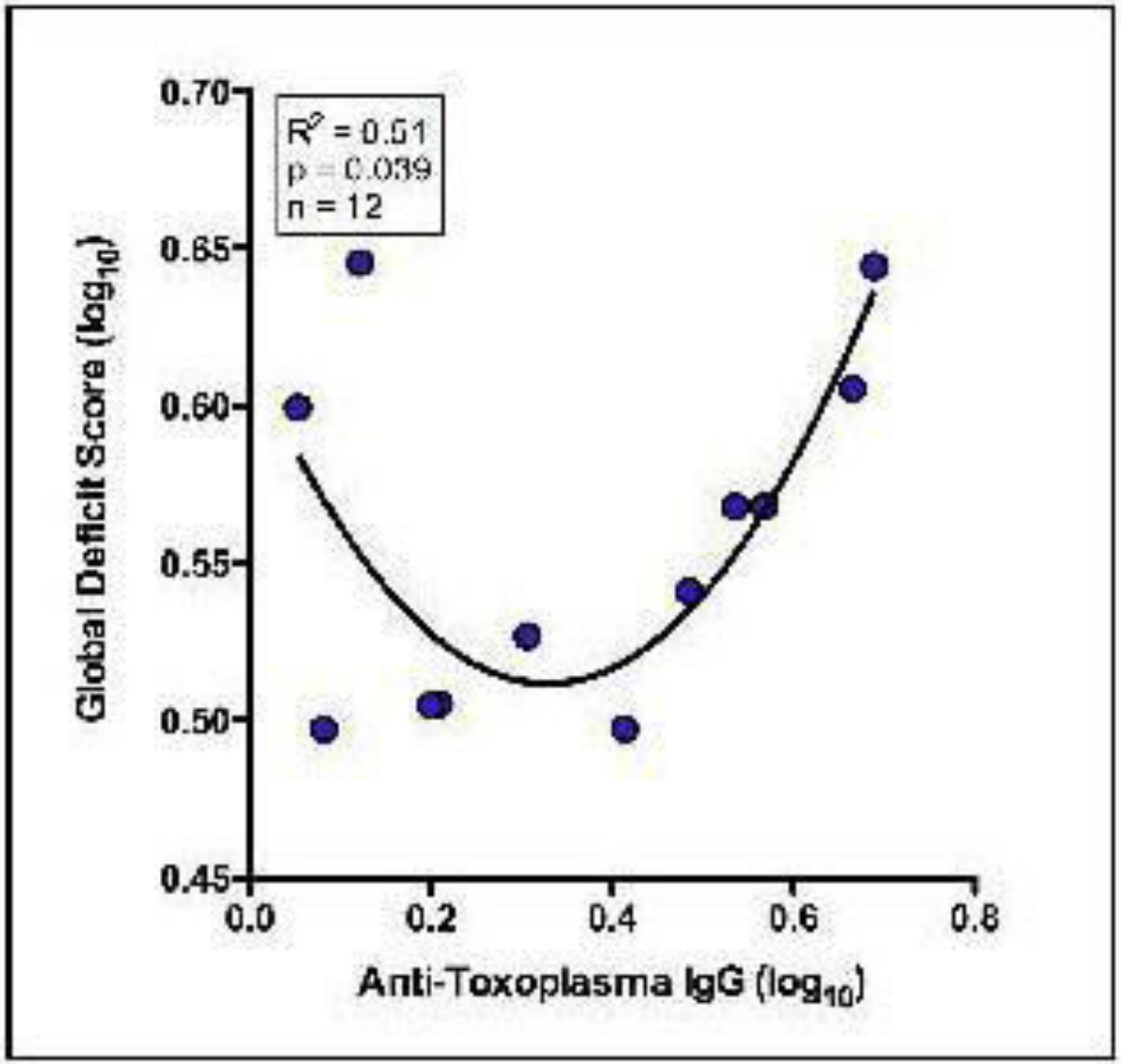
Latent toxoplasmosis and cognition

- Impaired cognition in seniors Gajewski, P. D., et al. (2014). Brain Behav Immun 36: 193-199; Dickerson, F., et al. (2014) J Nerv Ment Dis 202(8): 589-593;
- Immediate rather than delayed memory impairment in older adults Mendy, A., et al. (2014). Brain Behav Immun.

- Affecting cognitive function in certain groups (Gale, S. D., et al. (2014). Parasitology: 1-9)$\longrightarrow$ significant interactions between latent toxoplasmosis and
  - the poverty-to-income ratio
  - educational attainment
  - race-ethnicity

Latent Toxoplasmosis – brain disturbances in HIV-infected subjects

- No significant
  Toxoplasma se
  et. all, JAIDS (2015) 68(1
  - high baseline pr
  - data collected f
- Older subjects
  towards worse
  higher anti-To
  with worse fu
  Conference. Washingtc



Objective

- We aimed to evaluate the possible contribution of latent infection with *T. gondii* on
  - neurocognitive performance
  - depression
  - suicidal risk
  - disturbances associated with frontal-subcortical circuitry damage
  - risk taking behaviours

  in a group of young adults with chronic HIV infection since childhood

Methods

Study participants

- 194 HIV+ participants in the Romanian HIV Pediatric cohort who were infected with HIV in their first years of life (early 1990s) by parenteral non-IDU route
- 51 HIV- age matched participants

Neurocognitive assessment

- Standardised battery of tests assessing seven cognitive domains
- Neurocognitive impairment (NCI) was estimated using the global deficit score (GDS) with a cut-off $\geq$ 0.5
- Individual test deficit scores, determined via demographically-adjusted T scores generated from a healthy population of Romanian young adults, ranged from 0 (T score of $\geq$ 40) to 5 (T score < 20)

Depression, psychiatric disorders and risk taking behaviors

- The **Beck II depression inventory**
  - 0–13 = minimal symptoms
  - 14–19 = mild depression
  - 20–28 = moderate depression
  - 29–63 = severe depression
- The **Frontal System Behaviour Scale** (FrSBe)
  - 3 sub-scales: apathy, disinhibition, and executive dysfunction.
- The **Risk Assessment Battery (RAB)** is a self-administered, multiple choice questionnaire, assessing needle sharing practices and sexual activity associated with HIV transmission.
- **MINI-International Neuropsychiatric Interview (MINI-Plus),** evaluated DSM-IV criteria for current/past major depression and current/past suicidal risk

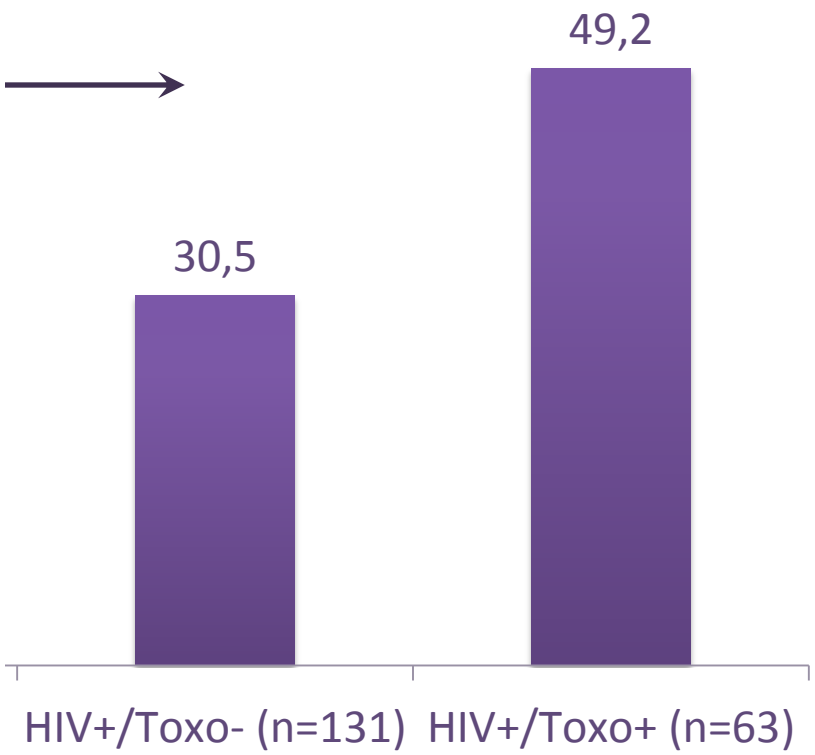
Characteristics of the participants

Characteristics of the participants	HIV-(n = 51)	HIV+(n = 194)
Male (n; %)	28; 54.9%	94; 48.4%
Age (mean; SD)	24.2(2.4)	24.0 (1.5)
Education**	13.3 (2.6)	12.1 (2.8)
HIV characteristics		
Time since estimated HIV transmission (years)[1]	-	23.7; 22.8-24.4
Time since HIV-diagnosis (years) [1]	-	14.8; 9.68-17.54
CD4 Current T-cells/µL[1]	-	**479**; 273-713
CD4 nadir cells/µL[1]	-	**93**; 22-190
Time since CD4 nadir years[1]	-	6.63; 1.57-11.46
AIDS defining diseases (n; %)	-	100 (51.5%)
HIV RNA in plasma undetectable (n; %)	-	118; 58.8%
ART characteristics		
Currently taking cART (n; %)	-	178 (91.7%)

	HIV-(n = 51)	HIV+(n = 194)
Toxoplasmosis characteristic		
IgG Toxo positive (n; %)	18; 35.3%	63; 32.4%
IgG Toxo IU/ml among positive participants	937; 232-1291	1090; 482-1604
Neurobehavioral characteristics		
GDS impaired (n, %)	6;11.7%	71; 36.5%
Depression		
BDI-II depression score >13 (n; %)*	1; 1.9%	23; 11.8%
Major depression diagnosis current	1; 1.96%	6; 3.09%
past	7; 13.72%	29; 14.94%
FrSBe (n, %)		
Apathy raw*	24.0; 6.1	26.6; 7.4
Disinhibition Raw Score	24.3; 6.6	25.4; 7.5
Executive dysfunction*	30.2; 8.2	33.2; 8.5
Total raw*	44.5; 13.2	48.5; 13.7
Suicidal risk (n; %)		
Current *	0; 0.0%	14; 7.2%
Past *	1; 1.9%	23; 11.8%

Latent Toxoplasma infection may result in increased cognitive difficulties in co-infected individuals

Anti-Toxoplasma antibodies were associated with a 60% increased relative risk of NCI (χ^2= 6.3, RR = 1.6, P=0.001).



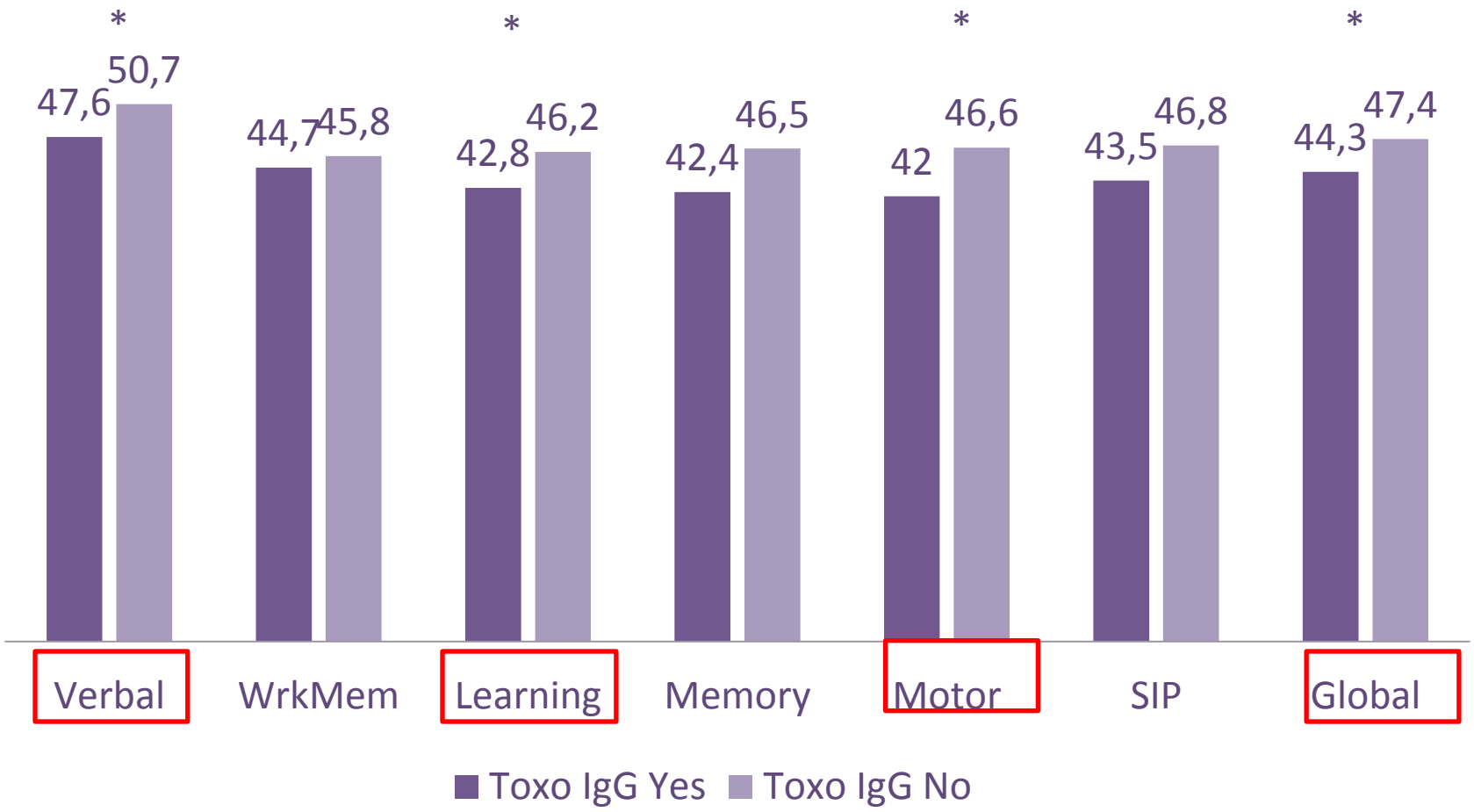
Multivariable models examining cognitive performance (GDS and T scores) using 2-way ANOVAS and logistic regression

	HIV-/Toxo-	HIV-/Toxo+	HIV+/Toxo-	HIV+/Toxo+	Effects	
GDS (sqrt)	0.20 (.21)	0.29 (31)	0.44 (.44)	0.59 (.48)	HIV***	Toxo*
Mean	50.5 (5.1)	48.7 (6.4)	47.3 (6.1)	44.8 (5.5)	HIV***	Toxo**
Executive	50.8 (6.5)	50.0 (7.1)	47.4 (7.2)	45.7 (7.6)	HIV**	
Verbal	50.1 (8.5)	47.8 (7.3)	50.3 (7.5)	47.7 (6.9)		Toxo*
Working Memory	50.0 (7.5)	50.3 (11.2)	45.9 (9.6)	44.3 (9.2)	HIV**	
Learning	50.7 (81)	49.0 (9.7)	46.3 (9.1)	43.2 (8.1)	HIV***	Toxo*
Memory	50.5 (7.5)	49.4 (10.5)	46.4 (9.8)	42.4 (10.2)	HIV***	Toxo**
Motor	49.5 (9.1)	48.8 (7.7)	46.4 (9.5)	44.3 (10.6)	HIV*	
SIP	50.9 (6.3)	47.2 (7.4)	47.2 (7.9)	44.2 (7.0)	HIV**,	Toxo **

Controlling for HIV

* $p < .05$, ** $p < .01$, *** $p < .001$

Effects of toxoplasma on NCI within the HIV+ group with undetectable HIV load (n=118)



*$p<0.05$

Relationship between Toxo status and risk behaviors

		HIV-/Toxo-	HIV-/Toxo+	HIV+/Toxo-	HIV+/Toxo+	p
FrSBe (raw score)	Apathy	24.1 (6.2)	23.8 (6.3)	26.8 (7.1)	26.1 (8.1)	0.14
	Disinhibition	24.4 (6.7)	24.1 (6.6)	26.1 (7.6)	23.9 (7.1)	0.20
	Executive	30.8 (7.9)	29.1 (8.9)	33.6 (8.7)	32.3 (8.0)	0.08
	Total	79.2 (18.5)	76.9 (20.2)	86.4 (20.7)	82.3 (20.5)	0.10
Drug use (% none)		96.7%	94.4%	96.2%	96.8%	0.87
Sex activity (median, IQR)		4 (2.5,5)	4 (2, 5)	2 (2,4)	2 (0,3)	<0.001*

Regression models:

- HIV & Toxo : no effect
- HIV : higher apathy (p=0.023), executive (p=0.019), and total (p=0.034)
- Toxo: no effect

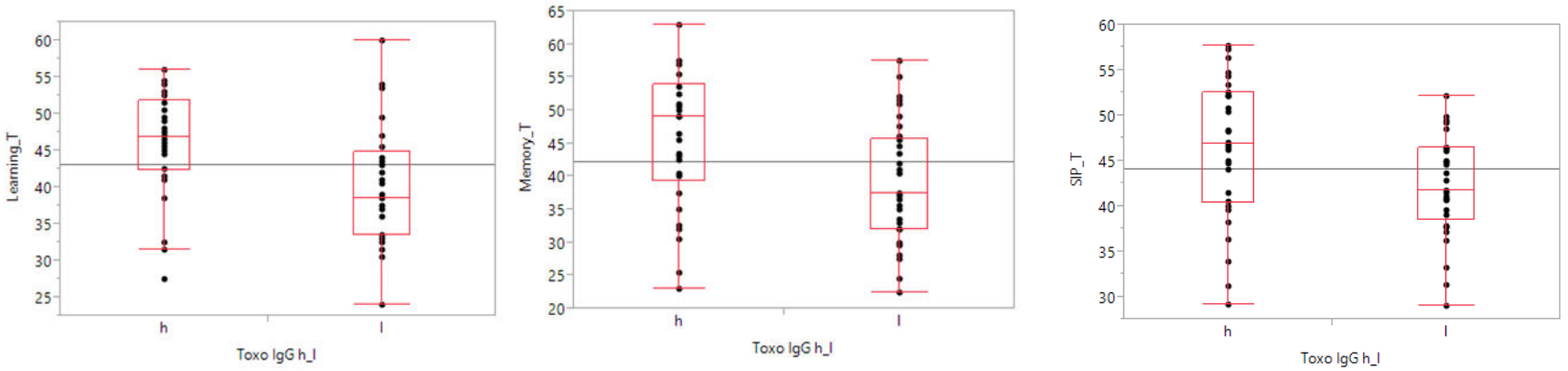
* HIV-ve participants had higher sexual risk-taking behaviours

No effect of latent toxoplasmosis

- current or past depression
- current or past suicidal risk

Based on MINI- Plus evaluation

Relationship between Toxo IgG levels cognitive performance & behavioral changes



These associations remained significant after controlling for medical covariates (AIDS, nadir CD4, years since diagnosis, and BMI) in multivariable models (all ps<0.03)

The group with **lower levels of Toxo IgG** had

- **higher** disinhibition (25.9 [7.8] vs. 21.7 [5.5], *p*=0.016)
- **higher** dysexecutive functioning(34.3 [9.0] vs. 30.1 [6.1], *p*=0.036)

- **higher** percentage of individuals with at least mild depression on the BDI (24.2% vs. 3.3%, *p*=.028).

Discussion (1)

1. Toxo and cognitive performances

- Detectable anti-Toxoplasma IgG antibodies were associated with a greater risk of NCI in HIV+ and HIV- participants

Latent Toxoplasmosis can be a potential confounder in attributing the cause of NCI to HIV

2. Latent Toxo infection was not associated with

- risk behaviours
- major depression
- suicidal thoughts

Discussion (2)

- Lower Toxo levels were associated with higher NCI rates and indicators of frontal systems dysfunction
  → **Toxoplasma may exert its negative effects as a result of slow and cumulative effects**
- Further studies and a longitudinal follow-up are warranted to determine the long-term impact of latent toxoplasmosis on NCI and also in behavioural changes and psychiatric conditions of HIV-infected patients

Acknowledgements

VBH team

Dan Duiculescu
Roxana Radoi
Ruxandra Burlacu
Gratiela Tardei
Simona Tetradov
Stefan Anton
Cosmina Gingaras

Anca Luca
Adina Talnariu (Bulacu)
Andreea Blaglosov
Adrian Luca
Silvia Suciu

Diana Sima

Study participants

IVN team

Simona Ruta
Aura Temereanca
Carmen Diaconu
Camelia Grancea

This work was supported by:
1R01MH094159
R21 MH0077487 P30
MH62512 from NIMH
and intramural funding from the HNRC International Core at UCSD

HNRC team

Cristian Achim
Tom Marcotte
Ron Ellis
Igor Grant
Davey Smith
Sanjay Metha
Scott Letendre

Terence Hendrix
Donald Franklin

Anya Umlauf
Reena Deutsch