Accelerated ageing in young adults infected with HIV since childhood ?

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Aging and HIV

- Prolonged survival in HIV infection is accompanied by an **increased frequency of non-HIV-related comorbidities which occur earlier** in HIV-infected patients than in individuals without HIV infection

- This **"accelerated aging"** appears to be largely related to **chronic inflammation**, chronic immune activation, and immunosenescence in HIV infection

- HIV infection - increased incidence of:
  - neurocognitive decline
  - CVD
  - malignancy
  - infection and chronic viral reactivation
  - osteoporosis
  - frailty

(Desquilbet L, et al, 2009)

Pathogenesis of HIVE in children

- HIVE **prevalence** in non treated children – **20-60%** *(Englund et al 1996, Foster 2006, Lobato et al 1996)*

- Although HAART has profoundly impacted the incidence of severe neurocognitive impairments, **HIV-infected children on suppressive regimens still experience neurocognitive deficits** *(Paramesparan et al, 2010)*
  - irreversible neuronal injury prior to initiation of ARV medications
  - neuronal injury from exposure to inflammatory responses
  - neurotoxic effects of the treatment itself
  - poor CNS penetration of ARV - ongoing CNS viral replication and/or inflammation not reflected systemically *(Heaton et al, 2011)*

- **CNS penetration effectiveness (CPE)** of a particular regimen *(Cysique LA et al, 2011, Eisfeld et al, 2013)*
- **"viral escape"** within the autonomous CNS compartment *(Tamula et al, 2003)*

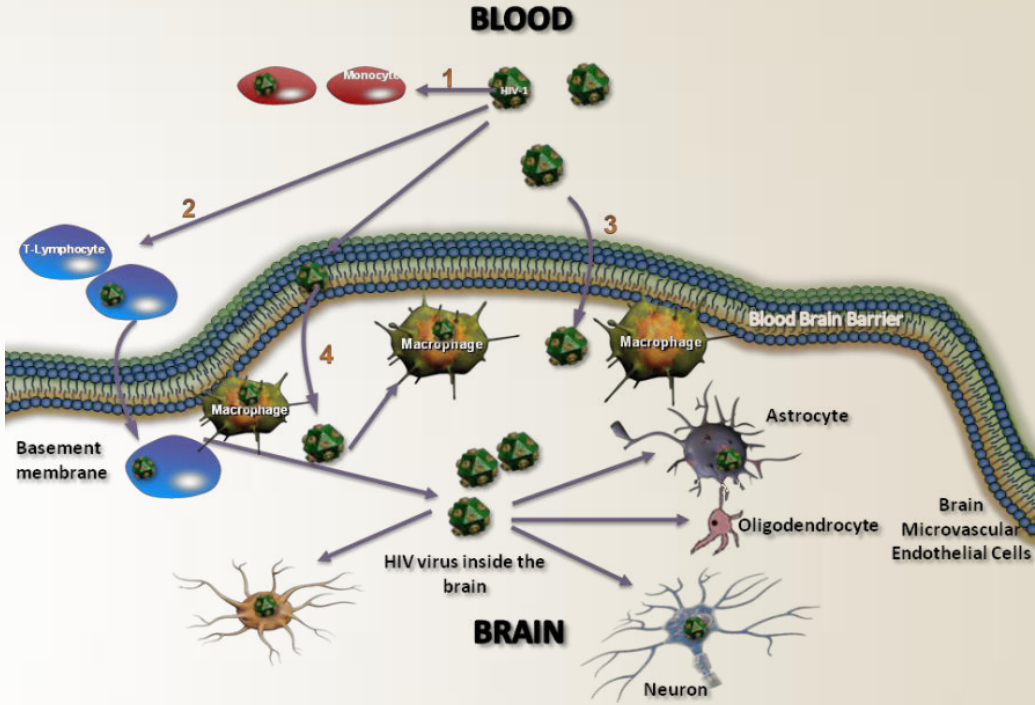
Pathogenesis of HIVE in children

- In children, **CSF HIV replication** is associated with neurocognitive impairment and HIVE severity - importance of early control of CSF virus *(Pratt et al, 1996, Sei et al, 1996)*
- Children require lifelong treatment → neurologic/ neurocognitive effects of HAART
- Risk factors associated with HIV-associated neurocognitive disorders in adults - vascular disease, premature neurodegeneration, or comorbidities such as hepatitis C *(Letendre et al, 2010, Wright et al, 2010)* - not known to be significant contributors to neurocognitive deficits in children and adolescents
- HIV-infected children now survive to adulthood → influence on existing neurocognitive deficits

Pathogenesis of HIVE in children - HIV neuroinvasion

- Spread of HIV into the CNS is thought to occur within weeks after infection *(An et al, 1999, Davis et al, 1992)*
- Structural changes are detectable by MRI within the first year *(Ragin et al, 2012)*
- Neuronal damage caused by HIV in the CNS is largely attributed to inflammatory responses

  (Kaul et al, 2001, Gonzalez-Scarano et al, 2005)
- Autopsy evidence suggests that neuroinflammation persists despite effective ART *(Anthony et al, 2005, Garvey et al, 2014)*



- **1) "Trojan Horse hypothesis" - via migration of infected monocytes which differentiate into perivascular macrophage**
- **2) The passage of infected CD4+ T cells into the brain**
- **3) The direct entrance of the virus via tight junctions across the membrane**
- **4) entrance of HIV-1 by transcytosis phenomenon**

Pathogenesis of HIVE
- differences in children vs adults

- The normal range of white blood cell counts in the newborn CSF is higher than in adults - leukocytes translocate more easily this barrier in newborns *(Greenlee JE, Carroll KC, 2004)*

- HIV infection in neural progenitor cells *(Schwartz L, Civitello L, Dunn-Pirio A, et al., 2007, Krathwohl MD, Kaiser JL, 2004)* and developing neurons *(Canto-Nogues et al, 2005)* in brain tissue from HIV-infected children has been described

- Neurotoxic effects of chemokines and HIV proteins may be of greater importance in the developing brain

Pathogenesis of HIVE
- neurologic/neurocognitive effects of HAART

- An early AIDS-defining illness increased the risk of chronic static encephalopathy during the preschool and early school age years *(Smith et al, 2006)*
- Treated HIV-infected children with no prior class C event - similar cognitive performance as their HIV-exposed uninfected peers *(Smith R et al, 2012)*
- Youth with HIV infection but no history of severe HIV disease exhibit low average to average cognitive performance similar to their HIV-exposed but uninfected peers *(Smith R et al, 2012)*
- Early treated infants have significantly better neurodevelopmental scores, compared with HIV-infected infants for whom treatment was deferred until clinical or immunological progression *(Laughton et al, 2012)*

CNS penetration effectiveness (CPE)

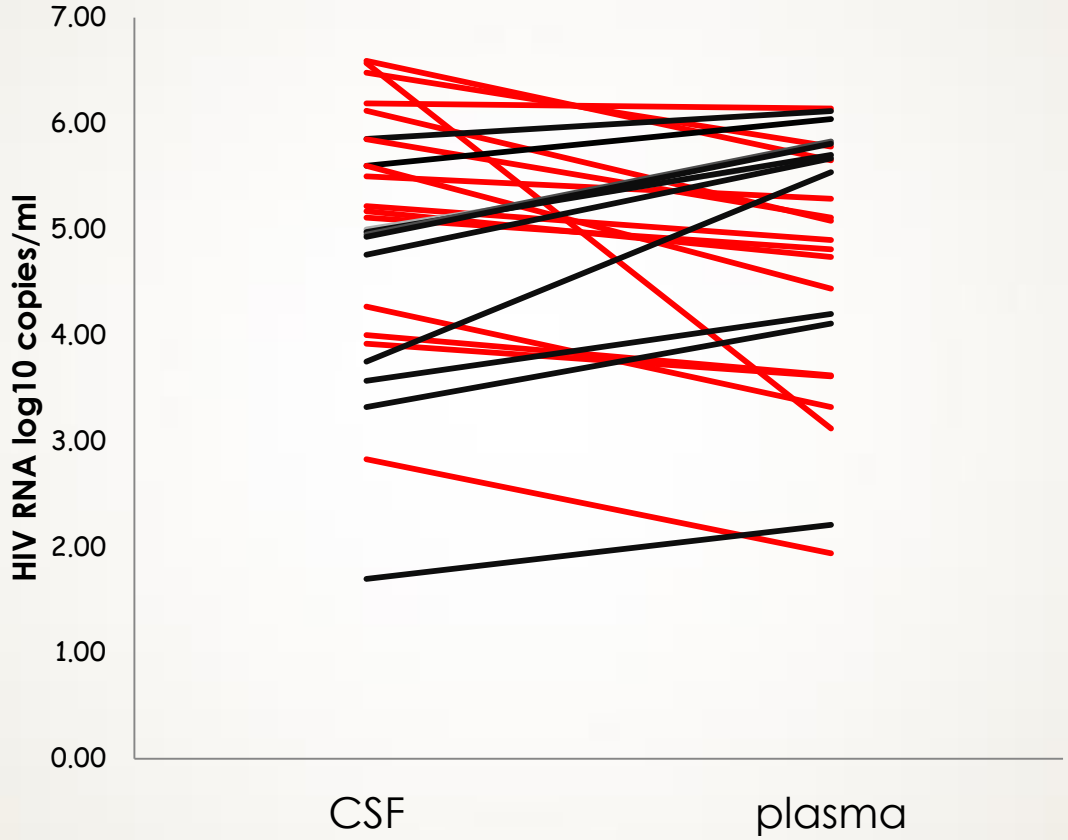
Pathogenesis of HIVE
- neurologic/neurocognitive effects of HAART

- Mechanisms of ART-associated neurotoxicity:
  - metabolic derangements associated with a variety of ARV medications *(Jayadev S, et al, 2009)*
  - mitochondrial toxicity associated primarily with nucleoside RT inhibitors *(Jayadev S, et al, 2009, Schweinsburg et al, 2005)*
  - proteasomal dysfunction associated with protease inhibitors *(Piccinini et al, 2005)*
  - and exacerbation of CNS vascular disease *(Mothobi et al, 2012)*
- Certain ARV drugs produce well-described neurologic side effects

Compartmentalization of HIV infection in the CSF - “viral escape”

- Neurocognitive deficits can emerge among HIV infected adults on ART with undetectable plasma viral loads
- Virus recovered from the CSF and plasma of individual patients is often genetically diverse *(Antinori et al, 2005, Liu et al, 2013, Canestri et al, 2010)*
- Adult patients on suppressive ART regimens with new or worsening neurologic symptoms not infrequently have detectable CSF viral load *(Canestri et al, 2010, Peluso et al 2012)*

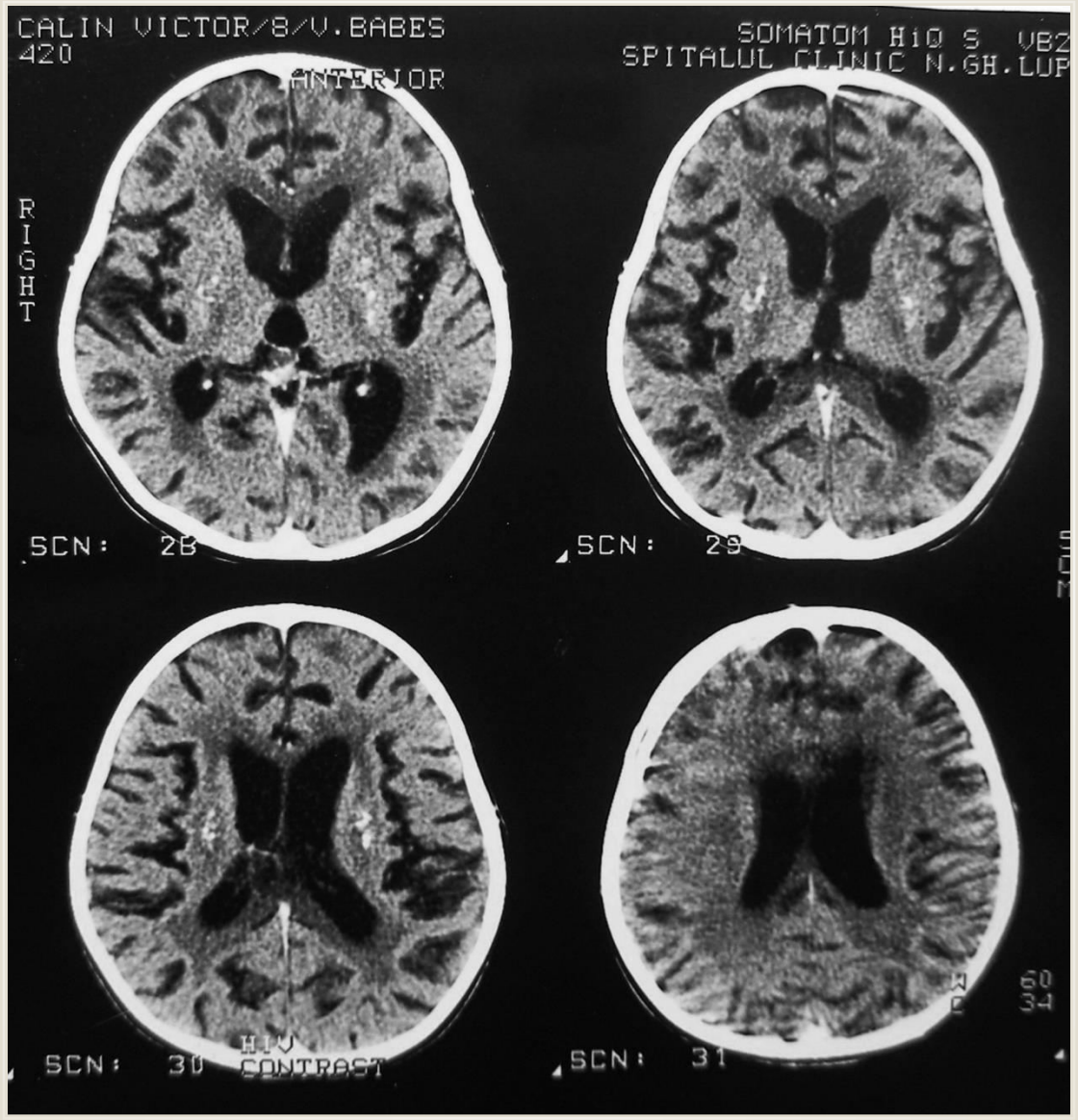
CSF and plasma levels in patients with HIVE from Romania



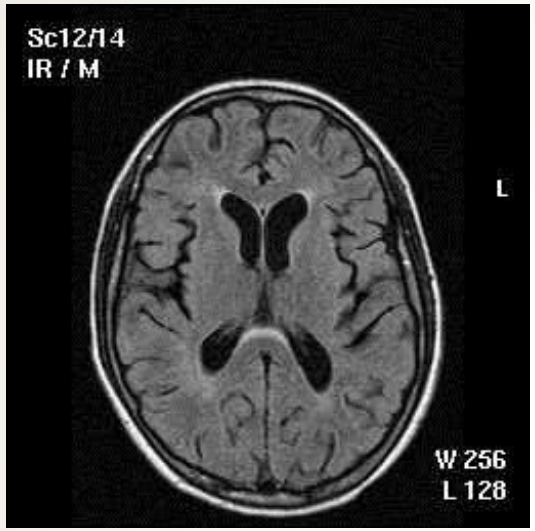
- **16 of 32 pts diagnosed with HIV encephalopaty had higher CSF levels**
- **3 of 11 patients with HIVE had altered BBB**

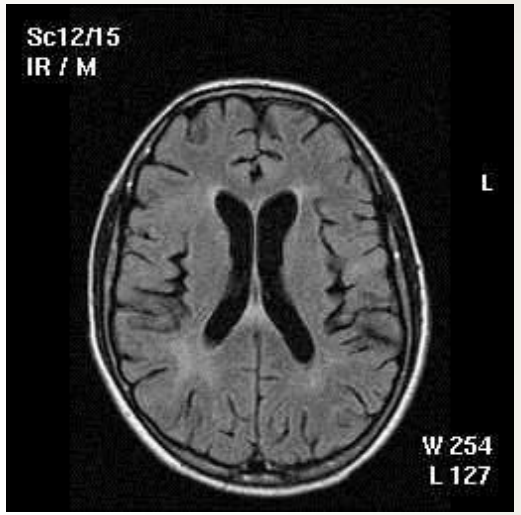
Duiculescu D et al IAS 2011 MOPE259

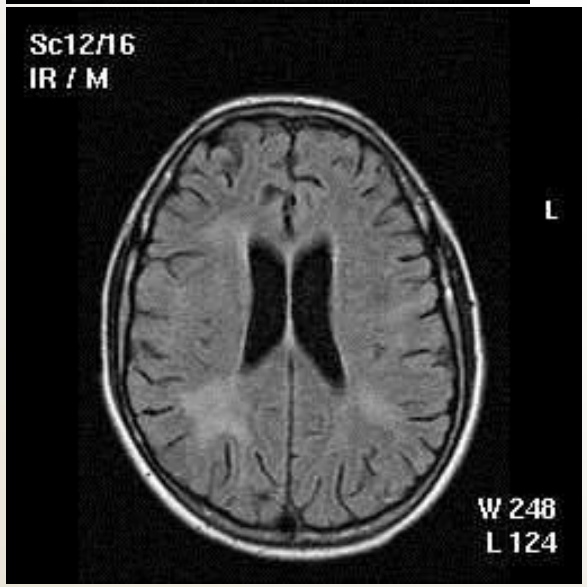
Brain CT – 8 y.o. child with cortical and subcortical atrophy and calcifications

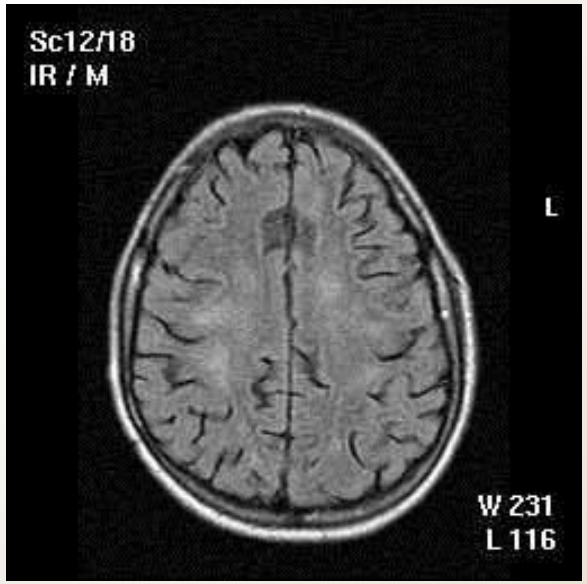


Demyelinating aspect and cortical atrophy in a 18 y.o with HIVE, cognitive deficit and paraparesis









Why is neurocognitive impairment important in children and adolescents?

- Neurocognitive impairment impacts
  - quality of life
  - school performance
  - risk behaviors
  - productivity in adulthood

- Neurocognitive impairment has practical implications
  - diminished ability to comprehend
  - diminished adherence to prescribed medication *(Ettenhofer ML et al, 2010, Malee K, et al, 2009)*

Challenges to describe neurocognitive impairment in adults infected with HIV since childhood

- Aim - to identify patients with more subtle deficits
- Detection of mild deficits needs more difficult tests that often take longer than the simple timed motor tests of the pre-ART era
- Global cognitive scores may overlook subtle deficits in one or more areas specific to PHIV children and may affect their performance on a different level *(Laughton B et al. 2013)*
- Additional reliable biomarkers with more pathophysiological validity are needed to transform this area of research

Adolescents with chronic HIV- infection

- Vertically infected
  - treated with cART
  - long-term non-progressors
- Horizontally infected - roughly comparable developmentally to their peers until late in their course *(Mitchell, W. (2001)*
  - Blood transfusion
  - Unsterilized needles

- Romanian pediatric cohort
  - Unique, homogenous
  - F-clade infection in the same period (1987-1990)
  - Similar genetic background
  - Similar length of exposure to ART (~15 years)
- Current age - 20-29 ans
- Sex ratio male/female=54/46
- HBV co-infection
- TB co-infection

General characteristics of the participants tested between 2012 and 2015

	HIV+ group n=222
Duration of infection years, date of infection based	**23.6 (22.8-24.6)**
Duration of infection years, diagnosis based	15.8 (10.2-18.3)
Plasma ND (<34 c/ml) (%)	**59.5%**
CSF ND (<34 c/ml) (%), n=72	86.6%
Current CD4 cell/ml, median (IQR)	479 (259-709)
Nadir CD4, median (IQR)	87.5 (22-190)
GDS imp (%)	35.05%

Active viral replication is associated with:

- male sex
- lower CD4 T-cell counts
- longer time on ART
- longer exposure to monotherapy

General characteristics participants

	2007-2009 (R21)		2012-2014 (R01)	
	R21 HIV- (n=20)	R21 HIV+ (n=49)	R01 HIV- (n=52)	R01 HIV+ (n=201)
Age	18.75 (1.02)	18.49 (0.77)	24.28 (2.44)	24.06 (1.52)
Education – mean years (SD) *	11.30 (.98)	9.78 (1.75)	**13.29 (2.60)**	**12.02 (2.81)**
Sex (% male)	60.00%	46.94%	55.7%	48.7%
Beck Depression Inventory-median (95% CI for median)	4.5* (3-6.8)	8* (6-13)	3 (1.7-10)	4 (1-15.5)
Unemployed/not in school°	0.2%	44.9%	34.15 %	66.48 %
GDS °	9.5%	48.9%	11.5%	36.3%
* p<0.05, ° p<0.001				

ART experience

ARV history % participants treated	Current use	**91.2**
	Past use	6.4
	Never used	2.4
Cumulative exposure to ARV (months)		129.8 (90.8-165.3)
Exposure to current regimen (months)		26.2 (10.6-47.5)
D-drug exposure	Past	48
	No	36.8
	Current	15.2

No correlates of deficit scores with:

- CD4: nadir, current, increase from nadir
- HIV RNA: zenith, current
- Previous AIDS
- ARV:
  - total exposure time to ARV's
  - exposure to current regimen
  - no of ARV's
- Depression
- Unemployment

Drug Abuse and HAND

- **Substance abuse - co-morbid condition** with HIV - additive or synergistic effects on the persistence and severity of neurocognitive dysfunction in patients with HAND (Martin-Thormeyer EM, et al , 2009)

- Cocaine, methamphetamine and opioid use - exacerbating the risk for **neuronal injury and neurocognitive impairment** in HIV+ patients *(Beyrer C, et al 2010, Byrd DA, et al, 2011, Dutta R, et al, 2012, The NSDUH Report: HIV/AIDS and Substance Use, 2010)*

- Methamphetamine has been linked to **increased neuroinflammation**, which may contribute to its neurotoxic effects *(Yamamoto et al. 2010; Clark et al. 2013).*

- Participants with histories of substance use (alcohol, cocaine, cannabis, opiates, methamphetamine) **did not have higher rates of neurocognitive impairment** or functional impairment in everyday life *(Byrd DA, et al, 2011)*

- The relative additive and synergistic effects of drugs of abuse on neuroinflammation in HIV+ individuals is not known

Drug use

Use > 5x (%)	HIV+	HIV-
alcohol	81.32	78.05
tobacco	50.55	51.22
marijuana	2.75	7.32
cocaine	1.10	2.44
methamphetamine	-	2.44
other stimulants	-	4.88
heroin	1.10	-
opioids	-	2.44
sedatives	0.55	-
anxiolitics	-	7.32
hallucinogens	0.55	2.44
dissociative drugs	-	2.44
popper	-	-
ecstasy	1.10	-
legal highs	2.04	5.00

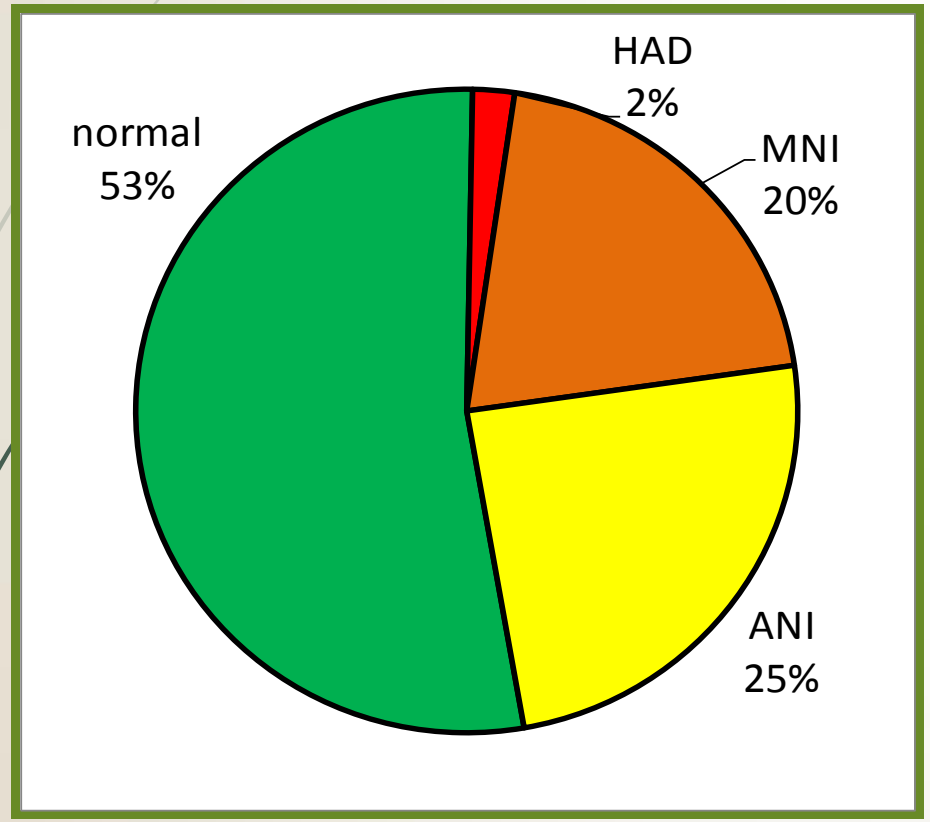
R01
4 controls
8 HIV + with > 1 drugs

Studies on NCI in children and adolescents with current good CD4 count

	Uganda (Ruel T et al, CID 2012:54: 1001-1009)	UK (Paramesparan Y et. al, JAIDS 2010: 55(1): 134-136)	Romania (Ene L et al EACS 2011, Abstract PS4/2)
Age	8.7	18.8	18.4
No of HIV+	93	6	49
Control group	HIV- age matched	Elderly HIV+ (65 yo)	HIV- age matched
Nadir CD4	N/A	393	86
Current CD4	655	619	517
Current HIV RNA	4.7 (4.2-5.1)	Undetectable in 4 of 6 pts	2.6 (1.6-5.8)
Subtype	A, D, C	N/A	F
Years of education		11.7	10.1
NCI impairment rates	Significant motor and cognitive deficits	67% (Cogstate)	47% (HNRC battery)

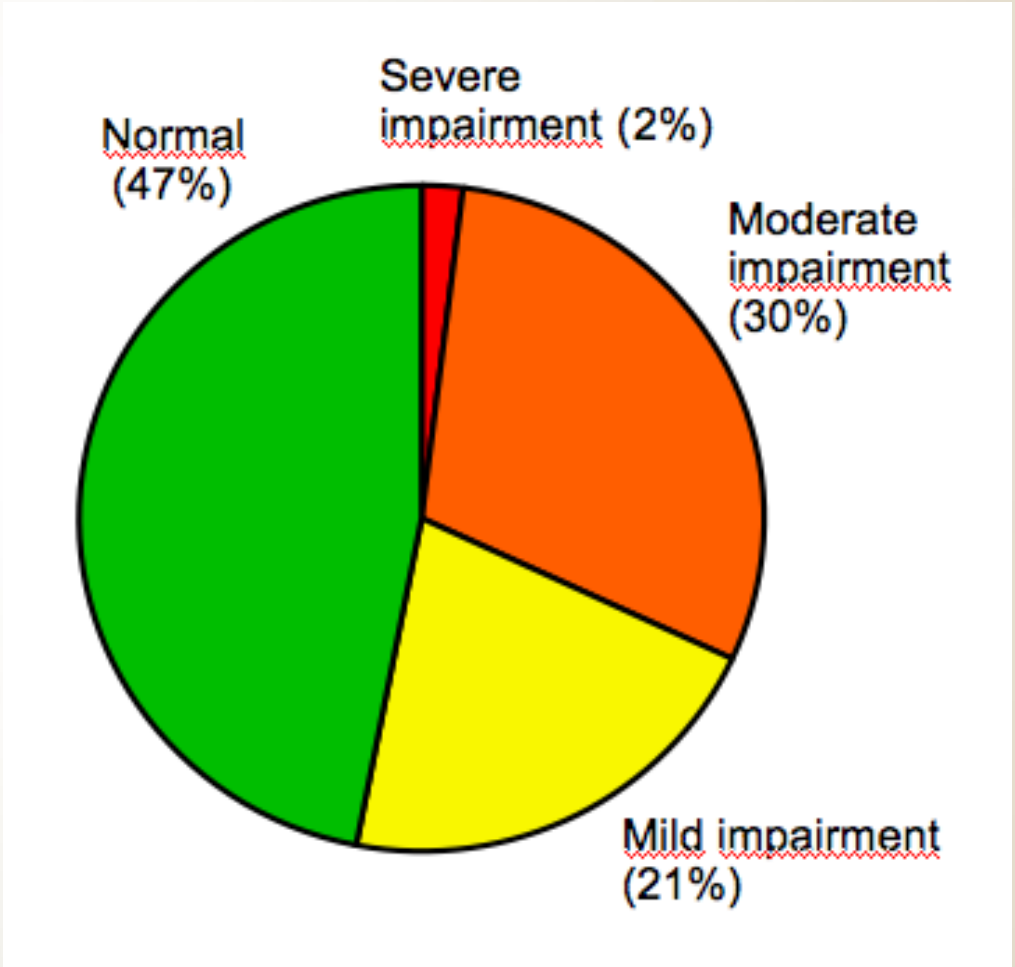
High overall prevalence of NCI in VBH population

VBH cohort



Duiculescu et al. 16th CROI 2009 # 477

CHARTER cohort



Heaton R et al., 16th CROI 2009

Systemic and plasma markers

- **Low CD4 nadir** → early treatment could substantially prevent the disorder *(Heaton et al. 2011, Ellis et al., 2011, Valcour et al., 2006, Lyons et al., 2011, Crum-Cianflone et al., 2013)*
- **Plasma-soluble CD14** - impairment in attention and learning *(Lyons JL , 2011)*
- **HIV DNA** circulating within mononuclear cells *(Valcour VG, et al., 2009, Shiramizu B, 2009, Valcour VG, 2010, Shiramizu B et al., 2012)*
- Increased trafficking of **activated monocytes** to the brain *(Lyons JL et al. , 2011, Pulliam L, Gascon R, Stubblebine M, McGuire D, McGrath MS., 1997, Kusdra L , 2002)*
- HIV affects the gut to potentially cause **microbial translocation** driving chronic inflammation, leading to HIV-associated dementia *(Ancuta P et al, 2008, Brenchley et al, 2006)*
- **Carotid intima–media thickness and glomerular filtration rate** were associated with performance speed on neuropsychometric tests, and intima–media thickness was also associated with memory impairment *(Becker JT et al, 2009)*
- Increased presence of **metabolic risk factors** *(McCutchan et al, 2012)*

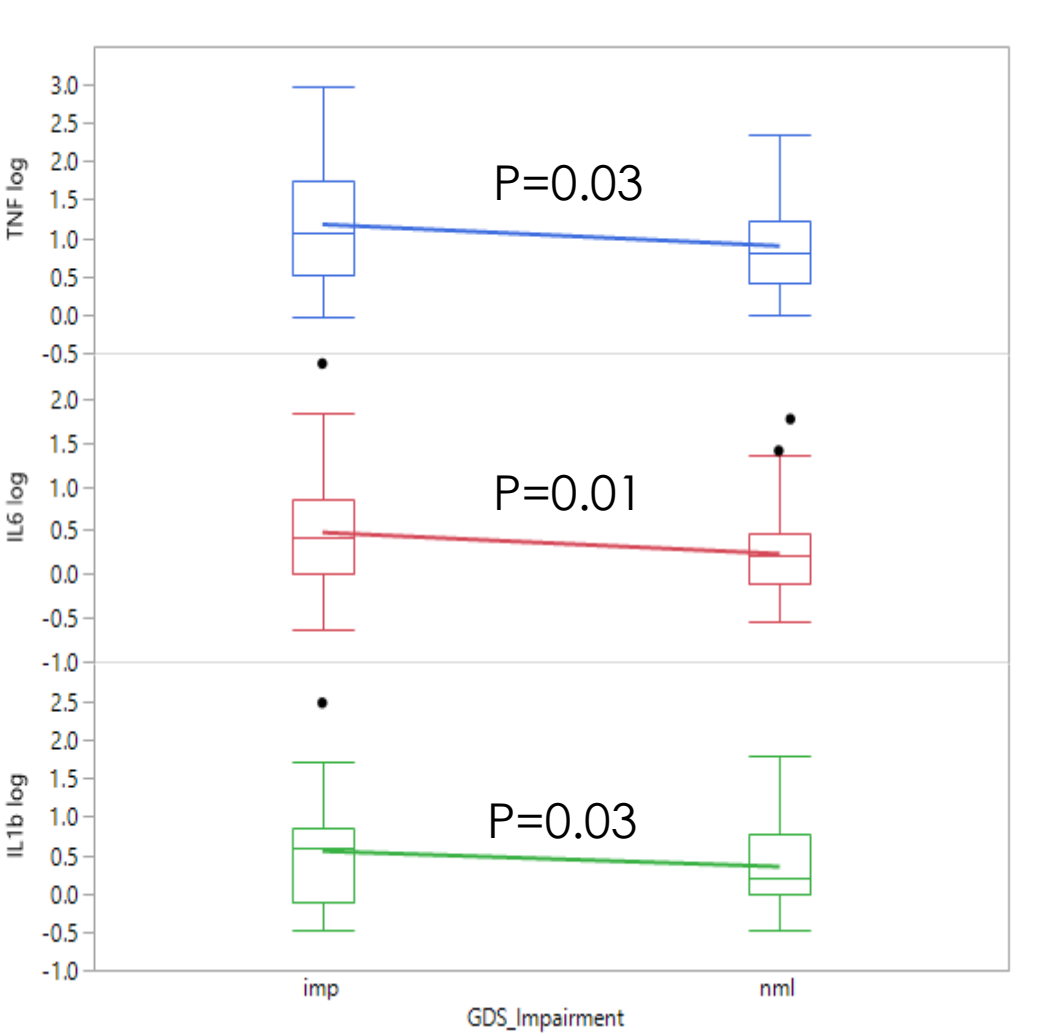
CSF markers

- Potentially identify patients at risk of HIV-associated neurocognitive disorder
- **Severity of cortical atrophy reflects the level of viral load in the CSF** *(Brouwers P et al, 2000)*
- **Virally suppressive ART protects against cortical neurodegeneration** (quantified by measuring microtubule-associated protein (MAP2) and synaptophysin (SYP) density in midfrontal cortex tissue sections) *(Bryant AK et al, 2015)*
- **Persistent immune activation markers** – IL 6, IL 8, and MCP-1, remain present in successfully treated patients *(Kamat et al, 2012)*
- **Markers of neuronal injury** (ex. neurofilament light protein) could also be associated with more advanced cognitive impairment *(Mellgren A, et al, 2007, Abdulle S, et al, 2007, Gisslén M, et al, 2007, Letendre S, et al, 2011)*
- Concentration of **tau protein** might be raised in HIV-associated dementia but not in ANI or MND *(Gisslén M, et al, 2007, Letendre S, et al, 2011, Ellis RJ et al, 1998, Clifford et al, 2009)*
- **MCP-1 and MMP-9** declined parallel with HIV RNA CSF load in children on ART *(McCoig C et al, 2004)*

Biomarkers

Plasma (n=144)	CSF (n=57)
IFN-γ	IFN-γ
IL-1β	IL-1β
IL-6	IL-6
IL-8	IL-8
TNF-a	TNF-a
Fractalkine	Fractalkine
IP-10	IP-10
MCP-1	MCP-1
	IL-10
	IL-12 p70

Neurocognitive impaired participants had higher IL1b, IL6 and TNFa levels

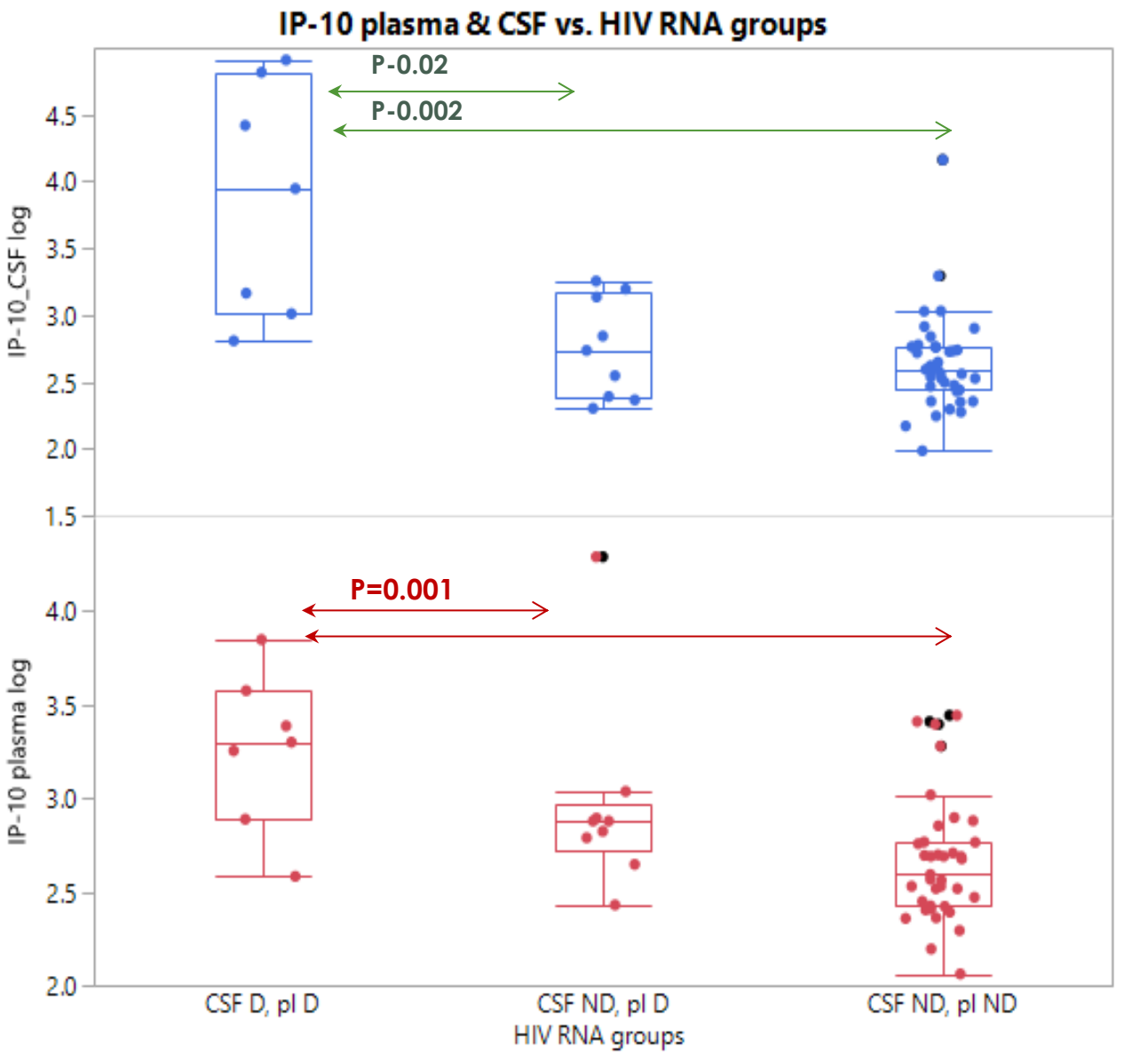


- TNF-a, sTNFR-II, and IL-6 have all been previously implicated in HIV disease progression *(Nixon and Landay 2010; Crowe et al. 2010, Achim et al. 1993; Mastroianni et al. 1990; Perrella et al. 1992; Vullo et al. 1995)*
- Lower sTNFR-II concentrations were associated with neurocognitive worsening and higher IL-6 concentrations were associated with neurocognitive improvement (Marcotte et al, 2013)
- TNF-a may link neurocognitive progression and remission

- SMART - elevated IL-6 at baseline and hs-CRP were significantly associated with mortality
- Greater age and body mass index were associated with higher IL-6 *Rodger AJ, 2009*

- **therapeutic implications?**

Correlation between IP 10 and HIV ARN levels



- G-CSF and IP-10 in plasma were significantly higher in HIV-impaired than HIV-normal cognition
- G-CSF, IL-8, IP-10 and MCP-1 in CSF showed significant difference between HIV-impaired and HIV-normal cognition group *(Yuan L et al, 2015)*
- 2 ACTG - higher IP-10 levels and higher MCP-1 levels correlated with lower cerebral metabolites in the brain regions considered
- higher levels of IP- 10 correlated with lower neuronal pattern scores and higher basal ganglia and inflammatory pattern scores, the same pattern which has been associated with (HAND) *Letendre et al, 2011*

Cardiac disease among HIV-infected children and adolescents

- In HIV-infected pregnant women treated with HAART - **no significant changes** in fetal cardiac parameters *(De la Calle M et al, 2015)*
- Significant burden of cardiac disease was seen among children with vertically-acquired HIV infection. Over **half** of asymptomatic 110 adolescents **had significant echocardiographic abnormalities** *(Miller R et al, 2012)*
- HIV-infected adolescents showed **higher intima-media thickness** *(Idris NS et al, 2014, Sainz T et al, 2015)*
- **Birth defects** - 39.34% *(Tudor AM, 2014)*
- **Comparable myocardial function and similar carotid intima-media thickness** *(Chanthong P et al, 2014)*
- **No differences** regarding cardiac abnormalities - vertically HIV-infected children and adolescents *(Sainz T et al, Pediatr Infect Dis J. 2015)*
- French Perinatal Cohort - specific **association between in utero exposure to ZDV and congenital heart disease** and a long-lasting postnatal myocardial remodeling in girls *(Sibiude J et al, 2015)*
- **Effects of HIV infection per se and antiretroviral therapy treatment?**

Cardiac disease among HIV-infected children and adolescents

- HIV-infected children have **higher levels of biomarkers of vascular dysfunction (sICAM, sVCAM, MCP-1, IL-6, and fibrinogen levels) than healthy children.** Risk factors associated with these biomarkers include higher waist to hip ratios and HIV disease severity *Millet Tr et al, 2010*

- Vertically HIV-infected **subjects on ART with no significant metabolic disturbances displayed increased sCD14 and soluble vascular cell adhesion molecule-1 (sVCAM) but not up-regulation of proinflammatory pathways** (C-reactive protein, interleukin-6, myeloperoxidase, monocyte chemoattractant protein-1, P-selectin and tissue plasminogen activator) *Sainz et al, 2014*

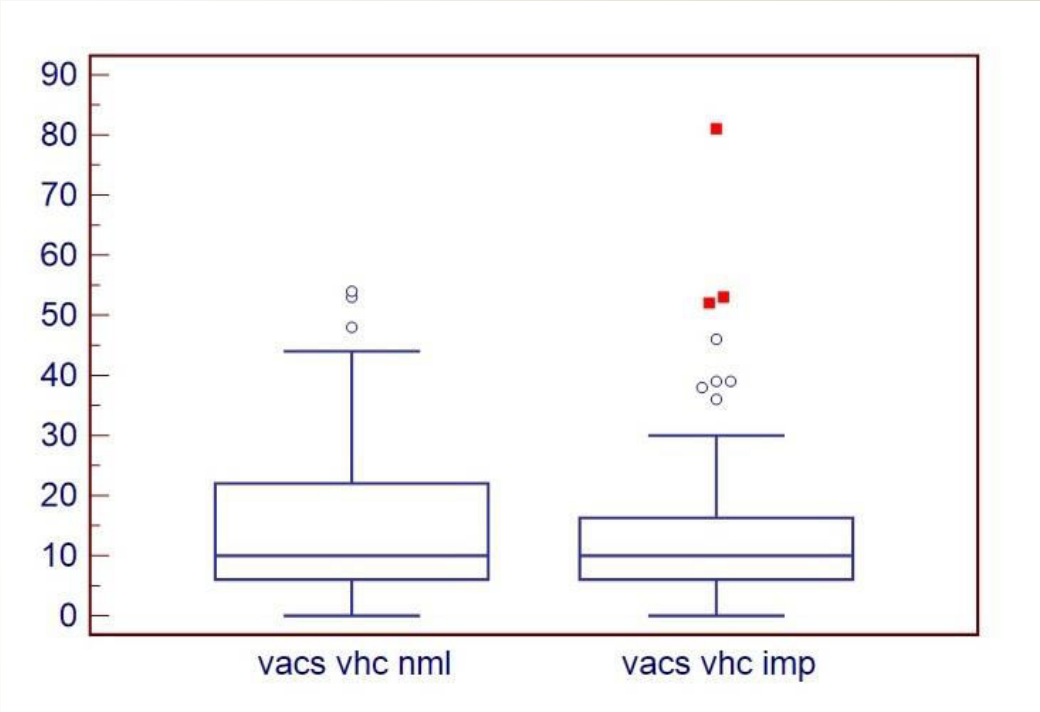
Metabolic risk factors didn't influence NCI

	HIV+ group n=201
Hemoglobin (g/dl) (N: 11-15) <11	13.8 (8.6-18.3)
Triglycerides (mg/dl) (N: 50-200) > 200	121 (41-1076) 16%
Total cholesterol (mg/dl) (N: 50-200) >200	173 (89-371) 59%
Albumin (g/l) (N: 35-50) <35	41 (29-56) 7.18%
Fasting blood sugar (mg/l) (N: <100mg/dl) >100	79 (57-144) 3.3%
AgHBS pos VHB DNA pos	29.2% 56%
AcVHC pos	2.2%
BMI > 25	9.45%
BMI <18.5	24.3%

Mortality index – VACS

- The Veterans Aging Cohort Study (VACS) index - 7 variables: age, CD4 count, HIV-1 RNA, hemoglobin, FIB-4, eGFR, and hepatitis C status – index for **5-year mortality** risk in HIV patients, with a **10-point increase** in the VACS score predicting a **10% increase in 5-year mortality** *(Justice AC et al, 2010, 2013)*

- Higher VACS Index scores were associated with concurrent **risk for global NCI** even when adjusting for psychiatric comorbidities for most cognitive domains in adjusted models.
- The VACS Index predicted concurrent NCI **beyond nadir CD4 and estimated duration of infection**
- Older age, lower hemoglobin, and lower CD4 counts were the VACS components most strongly linked to NCI (*Marquine MJ et a, 2014)*



- In our cohort, VACS index wasn't associated with a higher risk for NCI
- Hepatitis B?
- Median 11.5 (0-54), n=187

Discussion and conclusions

- We found a high prevalence of neurocognitive impairment – neurotropism of clade F?
  - Irreversible brain injury prior to initiating treatment
  - Persistence of low-level HIV replication in the brain
  - Persistence of inflammation and immune activation in the brain
  - Possible neurotoxicity of antiretroviral therapy on a developing brain
- No classical/metabolic risk factors associated with NCI
- Few confounders in terms of drug exposure, HCV coinfection, depression or psychiatric conditions in this group which may interfere with NCI
- NCI didn't correlate with HIV markers (CD4, HIV RNA)
- Challenges to assess NCI were related to difficulties in assessing functionality, to find an education matched control group
- The rates of NCI seem to decline on longitudinal follow-up

Future directions

- Further analyse longitudinal data (CD4, HIV RNA) and potential correlation with NCI

- New challenges for further studies emerge from this cohort:
- Cardiac disease evaluation
- Identify new risk factors
- What is the clinical significance of the high HBV coinfection for the CNS?
- Neuroimaging and MRS
- Follow up

A functional cure for HIV infection will need the virus to be silenced in all body compartments, including the brain *Valcour V, Sithinamsuwan P, Letendre S, Ances B. Curr HIV/AIDS Rep 2011*

Thank you!

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