HIV associated neurocognitive disorder (HAND) has become a common comorbidity in more than 30% patients infected by human immunodeficiency virus (HIV)-1, despite the presence of different antiretroviral treatments (ART). There are several clinical HAND syndromes depending on the severity of neurodegeneration: HIV associated dementia (HAD), mild neurocognitive disorder (MND) and asymptomatic neurocognitive impairment (ANI). HAND is associated with neuroinflammation and several biomarkers have been described throughout the last years to diagnose HAND, but a complete diagnostic analysis has not been found yet.

**HYPOTHESES**

The levels of molecules present in blood and cerebral spinal fluid (CSF) are associated to chronic inflammation. They can be used as biomarkers to predict neurodegeneration in HIV-1 infected individuals in order to treat the patients earlier. Furthermore, these biomarkers can be used to monitor treatments as nasal administration of anti-inflammatory drugs to improve symptoms of HAND.

**OBJECTIVES**

1. To find biomarkers associated with neurodegeneration in HIV-1 infected individuals
2. To find a transcriptomic profile associated with neurodegeneration in HIV-1 infected individuals
3. To test the panel of HAND biomarkers in mice treated with anti-inflammatory drugs administered intranasally

**MATERIALS AND METHODS**

**OBJECTIVE 1**

**PEOPLE RECRUITMENT AND SAMPLE COLLECTION (9 MONTHS)**

- **200 HIV+ ART, <20 copies/ml**: 65 HAND, 20 HAD, 20 MND, 25 ANI + 135 control
- **200 HIV-**: 100 neurodegeneration + 100 control
- Inclusion criteria check
- Diagnosis of HAND (HIVDS test)
- HAND questionnaire (AAN and HNRC criteria)
- Informed consent delivery

**ANALYSIS OF BIOMARKERS (3 MONTHS)**

- **Systemic biomarkers**: HIV viral load, CD4 nadir, CD4 count, CD4/CD8 ratio
- **Inflammation biomarkers**: IL-6, CRP, TNFα, IL-16, MCP-1
- **Monocyte/macrophage biomarkers**: sCD14, sCD163

**STATISTICAL ANALYSIS (3 MONTHS)**

- T student test
- Wilcoxon rank sum test
- Turkey test p<0.05

**OBJECTIVE 2**

**RNA PREPARATION (3 MONTHS)**

- 200 HIV+ ART, <20 copies/ml
- 200 HIV- (low MND ANI >30% MND) 5 HIV+ 20 HAD
- Pre ART era 20% HIV+ 100 HAND 20 HAD
- ART era 5 HIV+ 20 HAD

**ANALYSIS OF DATA (3 MONTHS)**

- Monocyte and CSF RNA-seq data
- Reads alignment
- Spliced Transcripts Alignment to Reference (STAR, Illumina)
- Trimm Galore software
- Differential expression
- Normalization of raw data
- Bioconductor software package p<0.05
- Neurodegeneration pathways relationship
- DAVID and STRING database
- qPCR of significant results found in RNA-seq

**OBJECTIVE 3**

**SCID MICE ORDER (3 MONTHS)**

- Ethics Committee approval
- 20 SCID mice (immunosuppressive model)

**HIV-1 MICE INFECTION**

- Intracerebral injections of HIV-1 monocyte-derived macrophages in 16 mice
- 16/20 HIV+ (HAND model) SCID mice
- 4/20 HIV- (no HAND model) SCID mice

**TREATMENT (9 MONTHS)**

- Intranasal BB-882 4 mice HIV+ BB-1101 HIV+
- Intraperitoneal and subcutaneous PBS 4 mice HIV+

**DATA ANALYSIS (3 MONTHS)**

- Monocyte and CSF RNA-seq data
- Reads alignment
- Identified differential expressed transcripts
- DAVID and STRING database
- qPCR of significant results found in RNA-seq

**EXPECTED RESULTS**

Some of the molecules analysed may be used as HAND biomarkers, considering that they are significantly more expressed in HIV+ individuals with HAND than in patients with no neurodegeneration.

**REFERENCES**


**DIFFUSION PLAN**

- 3 or more publications in high-impact journals of this area
- Results presentation at some relevant conferences, e.g. the Conference on Retroviruses and Opportunistic Infections (CROI)
- Dissemination of results to HIV seminars
- Dissemination of results to patients