Poster #506

DNA Methylation Changes in HIV-positive Men with Cognitive Decline

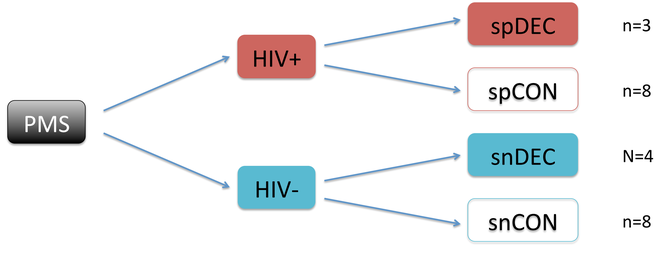
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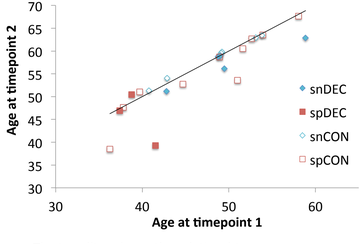
Background

- A subset of aging individuals with chronic, treated, HIV infection develops neurocognitive decline.
- Epigenetic factors, such as DNA methylation, have been implicated in the aging process and the development of chronic diseases.
- We hypothesize that long-term treated HIV infection has had a detectable impact on DNA methylation in these individuals, and this epigenetic change is associated with their cognitive impairment.
- We examined longitudinal changes in the whole genome methylation profile of 23 MSM enrolled in the Pitt Mens Study (PMS), a member study of the Multicenter AIDS Cohort Study (MACS)

Study participants and methods

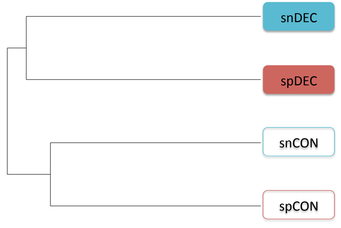


- Neuropsychological (NP) evaluation in the PMS includes measures from multiple cognitive domains related to the classification of HIV-associated neurocognitive disorder (HAND) [Becker *et al* 2014].
- Used to classify PMS participants as: seropositive with cognitive decline (spDEC); seropositive controls without decline (spCON); seronegative with cognitive decline (snDEC); seronegative controls without decline (snCON).
- PBMC pellets were obtained from the PMS repositories for each man, collected as close to ten years apart as possible.
- Genomic DNA was extracted from each pellet, and the DNA methylation profile was determined using the Illumina HumanMethylation450 array at the University of Pittsburgh Genomics Research Core.



The trendline shows the relationship
[Age at timepoint 2] = [Age at timepoint 1] + 10 years

DNA methylation differences between groups

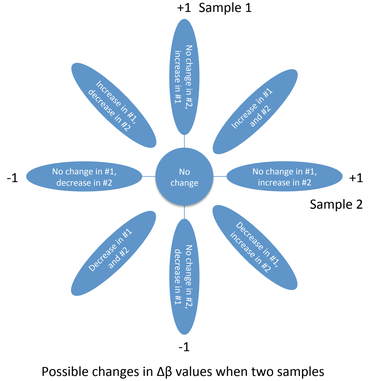


- Initial analysis of normalized data processed through Illumina's GenomeStudio software indicated a closer grouping between samples based on the presence or absence of cognitive decline than on HIV serostatus.
- Suggested to us that DNA methylation may be valuable as a biomarker of cognitive changes seen in both seropositive and seronegative individuals.

DNA methylation changes over time

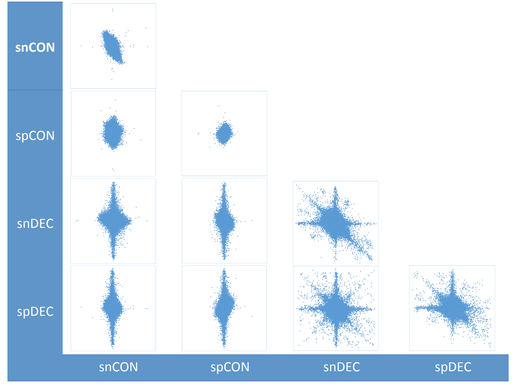
- Further analysis of methylation patterns using the R/Bioconductor package RnBeads [Assenov et al, 2014] did not reveal any methylation regions (*e.g.* genes, promoters, shores, shelves) where the amount of methylation differed consistently between our spDEC, snDEC, spCON, and snCON groups.
- Instead, we took advantage of the unique longitudinal nature of our dataset to measure the extent of change in methylation that occurred at each of the 485,000 sites contained in the Illumina HumanMethylation450 array in the time interval between the collection times for each person in our dataset.
- DNA methylation is known to change over time, and have been used to determine the "biological age" of an individual [Horvath 2013].
- Disorders of aging, including neurocognitive decline, are becoming more widespread in chronically HIV-infected individuals, such as those in the MACS.

- DNA methylation is measured as a β-coefficient, which ranges from 0 (fully non-methylated) to 1 (fully methylated) at any one timepoint.
- The difference in methylation between two timepoints $\Delta\beta = \beta_{Timepoint2} - \beta_{Timepoint1}$ can range from -1 to +1.
- Comparison of average Δβ values for the snDEC, spDEC, snCON, and spCON groups does not show any significant differences (data not shown) but pairwise comparisons of individual Δβ profiles for each sample reveals a clear differences.



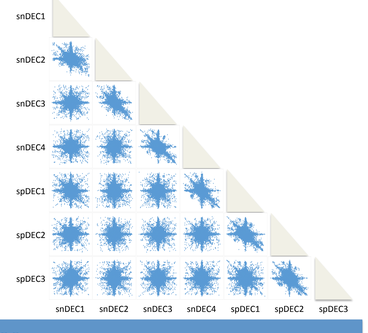
Possible changes in Δβ values when two samples are compared

Pairwise comparisons of DNA methylation change



- The presence of neurocognitive decline is associated with detectable changes in the methylation profile over a ten-year period.
- Individuals without decline – both seropositive and seronegative – do not show such changes
- HIV infection *per se* is not associated with this change, although we note that the average age of the spDEC cases in our study is 15 years younger than the snDEC cases.

- Pairwise comparisons of each snDEC and spDEC individual (right) show that this perturbation is consistently associated with cognitive decline.
- Similarly, pairwise comparisons of snCON and spCON individuals show a lack of perturbation (data not shown)
- The specific sites affected vary between individuals, and so there is no consistent pattern that can be detected in comparisons between groups.
- The cause of this chaotic change is unknown, and needs to be investigated further in larger numbers of seropositive and seronegative individuals with and with out cognitive decline.



References

- Assenov Y, Müller F, Lutsik P, Walter J, Lengauer T, Bock C (2014) "Comprehensive Analysis of DNA Methylation Data with RnBeads." Nature Methods, **11**:1138-1140.
- Becker JT, Kingsley LA, Molsberry S, Reynolds S, Aronow A, Levine AJ, Martin E, Miller EN, Munro CA, Ragin A, Sacktor N, Selnes OA (2014) "Cohort profile: recruitment cohorts in the neuropsychological substudy of the Multicenter AIDS Cohort Study." Int J Epidemiol [epub ahead of print, PMID: 24771276].
- Horvath S (2013) "DNA methylation age of human tissues and cell types." Genome Biol **23**:R115.

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