Biomarkers of ageing in HIV-positive individuals and matched controls

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Background

Despite successful combination antiretroviral therapy (cART), HIV+ve people have been suggested to experience signs of more rapid ageing compared to similarly aged controls.

Aims

- To compare established ageing biomarkers in HIV+ve people aged ≥45 years and comparable HIV-ve controls.
- To identify factors associated with apparent age advancement in both groups.

Methods

The COBRA collaboration

The COBRA cohort study investigates the potential link between HIV and age-associated comorbidities in a cohort of older HIV+ve individuals on cART and otherwise comparable HIV-ve controls in the UK and the Netherlands.

134 HIV+ve individuals aged ≥45, on cART and with a plasma HIV viral load <50 copies/mL for ≥12 months prior to enrolment were recruited at HIV outpatient clinics; **79 similarly aged HIV-ve controls** with comparable socio-demographic and lifestyle characteristics, were recruited from sexual health centres.

Age advancement

The biological age of each individual was derived using a set of 10 biomarkers (see box) identified through the EU FP7 MARK-AGE project (www.markage.eu) and measured in blood cells (B), plasma (P) or serum (S).

They were selected as best predictors of chronological age among approximately 400 candidate biomarkers and combined using a set of weights.

- cumulative level of cytosine methylation at gene positions (B):
  - ELOVL Fatty Acid Elongase 2 CpG 15,16,17
  - ELOVL Fatty Acid Elongase 2 CpG 11,12,13,14
  - FHL 2 CpG 11,12
  - FHL 2 CpG 13,14,15
  - FHL 2 CpG 16,17
- Dehydroepiandrosterone sulphate (P)
- N-glycan peak 6 (S)

only male subjects:
- Alpha-2 macroglubulin (S)
- Lycopene (P)
- Prostate specific antigen (S)

only female subjects:
- Ferritin (S)
- N-glycan $\log_{10}$(peak 1/peak 6) (S)
- a-Tocopherol (S)

Age advancement for each individual was defined as the difference between biological and chronological age.

Statistical analysis

- Age advancement in each group was assessed for significance using the one-sample t-test and the difference between the two groups using the unpaired t-test.
- Associations between age advancement and HIV status, lifestyle, viral [cytomegalovirus (CMV), chronic hepatitis B (HBV) and C (HCV) viruses] and HIV parameters were investigated using t-tests, Pearson's correlation coefficient (rho) and linear regression.

Results

Participant characteristics (Table 1)

Median (IQR) or n (%)	HIV+ve	HIV-ve	*p*
Age [years]	56 (51, 62)	57 (52, 65)	0.27
Male	125 (93%)	73 (92%)	0.79
Black-African	16 (12%)	2 (3%)	0.03
MSM	104 (78%)	59 (75%)	0.45
Years of education	14 (13, 16)	16 (14, 17)	0.23
Smoking status (self-reported)			0.24
Current smoker	40 (30%)	20 (25%)	
Ex-smoker	58 (43%)	29 (37%)	
Alcohol consumption (self-reported)			0.04
Current drinker	104 (78%)	71 (90%)	
Previous drinker	18 (13%)	3 (4%)	
Recreational drug use (past 6 months)	44 (33%)	18 (23%)	0.16
Chronic HBV infection	7 (3%)	0 (0%)	0.05
Chronic HCV infection	5 (2%)	0 (0%)	0.08
CD4 count [cells/µL]	618 (472-806)	N/A	
Years since HIV diagnosis	15.0 (9.1-20.0)	N/A	
Prior AIDS	42 (31.3%)	N/A	
Nadir CD4 count [cells/µL]	180 (90-250)	N/A	

Table 1 Socio-demographic and lifestyle characteristics of study participants [MSM: men who have sex with men; IQR: interquartile range].

Age advancement in HIV+ve and HIV-ve individuals

Biological age was significantly greater than chronological age by **13.2 (95% CI: 11.6, 14.9)** years in the HIV+ve group and by **5.5 (3.8, 7.2)** years in the HIV-ve group (p<0.001 for each) with a significant difference between the two groups (p<0.001).

Age advancement and lifestyle factors (Table 2)

No significant associations were found between age advancement and either gender or lifestyle factors in HIV+ve and HIV-ve individuals.

		Mean (95% CI) age advancement	Difference (95% CI) between groups	*p*
Gender	*Male*	10.3 (8.9, 11.7)	0	0.97
	Female	10.4 (6.1, 14.7)	0.1 (-5.1, 5.2)	
Smoking status	*Never smoked*	9.4 (7.1, 11.6)	0	0.34
	Current smoker	10.1 (8.1, 12.0)	0.7 (-2.4, 3.8)	
	Ex-smoker	11.8 (9.0, 14.6)	2.5 (-1.0, 5.9)	
Alcohol consumption	*Never drinker*	11.8 (8.2, 15.3)	0	0.43
	Current drinker	12.6 (8.7, 16.4)	0.6 (-5.6, 7.2)	
	Previous drinker	10.0 (8.5, 11.5)	-1.8 (-6.8, 3.2)	
Recreational drug use	*No*	10.6 (7.3, 13.9)	0	0.90
	Yes	10.1 (8.3, 11.9)	-0.5 (-4.3, 3.3)	

Table 2 Association between gender, lifestyle factors and age advancement in HIV+ve and HIV-ve individuals.

Age advancement and viral co-infections (Table 3)

Higher total and high avidity anti-CMV IgG antibody titer were both associated with an increased age advancement, independently of HIV-status (rho=0.24, p=0.03 and rho=0.25, p=0.02, respectively). Chronic HBV, but not HCV, co-infection was associated with greater aged advancement in HIV+ve individuals.

		Mean (95% CI) age advancement	Difference (95% CI) between groups	*p*
Chronic HBV co-infection	*No*	12.8 (11.1, 14.5)	0	0.01
	Yes	22.0 (10.5, 33.4)	9.1 (1.9, 16.4)	
Chronic HCV co-infection	*No*	10.2 (8.9, 11.5)	0	0.36
	Yes	14.2 (3.7, 24.7)	4.0 (-4.6, 12.7)	

Table 3 Association of HBV and HCV co-infections with age advancement in HIV+ve individuals.

Age advancement and HIV parameters (HIV+ve cohort only)

In HIV+ve persons, a positive correlation was found between age advancement and time since HIV diagnosis, duration of cART and time with a CD4 count <200 cells/µL (Figure 1). Due to the strong correlations between these three measurements, it was not possible to separate their effects in multivariable models.

Figure 1 Regression coefficient (with 95% CI) of HIV parameters demonstrating estimated associations with age advancement in univariate regression models.

Conclusions

- **Both successfully treated HIV+ve individuals and comparable HIV-ve controls show signs of age advancement, but this is significantly greater in HIV+ve people.**
- Total and high avidity anti-CMV IgG antibody titer and HBV (but not lifestyle factors) seem to be associated with increased age advancement. Longer exposure to HIV, cART and/or a low CD4 count appear to be associated with increased age advancement, although as all three markers were closely correlated we were unable to identify which of these was most strongly associated with age advancement.
- The effects of HIV on monocytes and lymphocytes, and unmeasured confounders may have contributed to the increased age advancement observed in HIV+ve people.

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