



IMPACT OF HAART INTERRUPTION ON PLASMA INFLAMMATORY MARKERS ASSOCIATED WITH CARDIOVASCULAR DISEASE. 24-MONTH RESULTS FROM A RANDOMIZED STUDY

Montserrat Olmo¹, Carlos Alonso-Villaverde², Maria Penaranda³, Felix Gutierrez⁴, Joan Romeu⁵, Maria Larrouse⁶, Pere Domingo⁷, Jose Antonio Oteo⁸, Jordi Curto¹ and Daniel Podzamcz¹ for the STOPAR team

¹Hospital Universitari de Bellvitge, Barcelona; ²Hospital Sant Joan de Reus, Tarragona; ³Hospital Son Dureta, Mallorca; ⁴Hospital General de Elche, Alicante; ⁵Hospital Germans Trias i Pujol, Barcelona; ⁶Hospital Clinic, Barcelona; ⁷Hospital Sant Pau i Santa Creu, Barcelona; ⁸Hospital de la Rioja, Logroño. SPAIN



BACKGROUND

HIV infection has been related to an increased level of pro-inflammatory markers [1]. HAART interruption has been associated with an increase in cardiovascular risk [2], being the probable mechanism the uncontrolled viral replication leading to an increase in several inflammatory proteins [3].

OBJECTIVE

To analyze the influence of CD4-guided HAART interruption on inflammatory markers associated with cardiovascular disease

PATIENTS AND METHODS

Design: Randomised, open, multicenter, 36 month completed trial
Setting: 7 Hospitals in Spain with extensive experience in HIV management
Patients: Adult HIV-infected patients on stable HAART for ≥ 6 months presenting undetectable viral load for ≥ 3months and more than 500 CD4 cells/mm3 for ≥ 6 months.

Intervention: After stratification by CD4 nadir (> or <200 cells/mm3) and NNRTI or PI use, patients were centrally randomised to continue treatment (CT) or to treatment interruption (TI). HAART was reinstated in TI-group if CD4 value decreased to <350 cells/mm3.

Follow-up: Clinical status and standard laboratory parameters were assessed at baseline, months 1, 2, 3 and every 3 months thereafter until month 36.
Primary endpoints: disease progression, death, CD4<200 cells/mm3 or virological failure after HAART reintroduction.

INFLAMMATORY MARKERS SUBSTUDY: Interleukin (IL)-6, IL-8, monocyte chemoattractant protein (MCP-1), soluble vascular cell adhesion molecule (s-VCAM), soluble P-selectin (sP-selectin), tissue plasminogen activator (tPA) and soluble CD40 ligand (sCD40L) were measured at baseline, month 12, 24 and 36 using a multiplex cytometric bead-based assay (Human Cardiovascular 7plex FlowCytometry Multiplex; BenderMedsystems GmbH, Vienna, Austria) and were analyzed on a EPICS-XL-MCL flow cytometer (Beckman Coulter; IZASA; Barcelona; Spain) following the manufacturer's protocol. TI-patients with at least 70% time off-HAART and CT-patients were analyzed and compared. MCP-1, s-VCAM and sP-selectin results are expressed as % of change from baseline. Logarithmic transformed values instead of absolute values of IL-6, IL-8 and sCD40L were used due to the skewed distribution shown by these variables, therefore their results are expressed as % of logarithmic change from baseline.

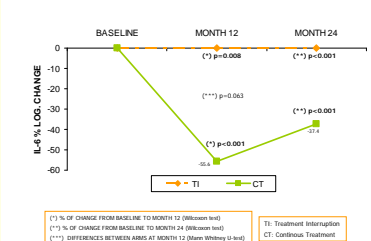
Follow-up: 24 month results are presented.
Statistical analysis: data are presented with median % of change from baseline and with median absolute values at each time point. Statistical tests used to compare between and within groups are shown under each table or graph, when appropriate.

STOPAR STUDY

BASILINE CHARACTERISTICS	CT (n=50)	TI (n=56)	TOTAL (n=106)	p-value
Gender, n (%)				
Male	40 (80.0)	38 (67.9)	78 (73.6)	0.189*
Female	10 (20.0)	18 (32.1)	28 (26.4)	
Age (years), median (min-max)	41 (26-81)	39 (24-71)	40 (24-81)	0.322*
Drug user	16 (32.0)	16 (28.6)	32 (30.2)	
Risk practice, n (%)				
Heterosexual	19 (38.0)	22 (39.3)	41 (38.7)	0.821*
Homosexual	14 (28.0)	15 (26.8)	29 (27.4)	
Other	1 (2.0)	3 (5.4)	4 (3.8)	
AIDS, n (%)				
No	47 (94.0)	53 (94.6)	100 (94.3)	1.000*
Yes	3 (6.0)	3 (5.4)	6 (5.7)	
Baseline regimen, n (%)				
NNRTI*	42 (84.0)	48 (85.7)	90 (84.9)	0.806*
PI†/‡	8 (16.0)	8 (14.3)	16 (15.1)	
Baseline CD4 (cells/mm ³), median (min-max)	813 (523-814)	873 (540-1817)	845 (523-817)	0.384*
Nadir CD4 (cells/mm ³), median (min-max)	301(176-720)	345 (105-1214)	327 (105-214)	0.660*

* Chi square test. † Nucleoside tase. ‡ Nucleoside tase. NNRTI: Non Nucleoside Reverse Transcriptase Inhibitor. PI: Protease Inhibitor. VL: Viral Load. ART: antiretroviral. ††: Abacavir, AZ: zalcitabine.

IL-6 % OF LOG. CHANGE BY TREATMENT ARM



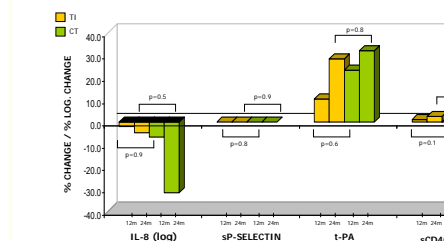
IL-6 MEDIAN ABSOLUTE VALUES (pg/mL)			
BASELINE	12 m	24 m	
TI	0.2	0.7	0.3
CT	2.7	0.2	0.3

INFLAMMATORY MARKERS SUBSTUDY

BASILINE CHARACTERISTICS	CT (n=44)	TI (n=33)	Total (n=77)	p-value
Gender, n (%)				
Male	36 (81.8)	24 (72.7)	60 (77.9)	0.341*
Female	8 (18.2)	9 (27.3)	17 (22.1)	
Age (years), median (min-max)	41 (28-64)	40 (27-64)	41 (28-64)	0.601*
Height (cm), median (min-max)	172 (151-183)	166 (151-183)	170 (151-183)	0.016*
Weight (kg), median (min-max)	69.0 (54.0-95.0)	69.9 (57.0-96.0)	69.1 (57.0-96.0)	0.912*
Drug user	15 (34.1)	10 (30.3)	25 (32.5)	
Risk practice, n (%)				
Homosexual	16 (36.4)	12 (36.4)	28 (36.4)	0.655*
Heterosexual	12 (27.3)	8 (24.2)	20 (26.0)	
Other	1 (2.3)	2 (6.1)	3 (3.9)	
AIDS, n (%)				
No	41 (93.2)	31 (93.9)	72 (93.5)	1.000*
Yes	3 (6.8)	2 (6.1)	5 (6.5)	
Baseline regimen, n (%)				
NNRTI	36 (81.8)	28 (84.8)	64 (83.1)	0.729*
PI/‡	8 (18.2)	5 (15.2)	13 (16.9)	
CD4 (cells/mm ³), median (min-max)	806 (523-1814)	877 (524-1817)	846 (523-1817)	0.104*
CD4 nadir, median (min-max)	296.5 (116-720)	416 (121-1214)	369 (116-1214)	0.091*
VL (copies/mL), median (min-max)	99 (19-199)	99 (19-199)	99 (19-199)	0.208*
T-Cholesterol (mmol/L), median (min-max)	5.0 (2.3-7.3)	5.1 (2.3-7.9)	5.1 (2.3-7.9)	0.216*
HDL-C (mmol/L), median (min-max)	1.2 (0.2-2.1)	1.4 (0.9-2.3)	1.3 (0.2-2.2)	0.504*
LDL-C (mmol/L), median (min-max)	2.9 (1.8-5.3)	3.1 (1.4-4.8)	3.0 (1.4-5.3)	0.379*
Triglycerides (mmol/L), median (min-max)	1.9 (0.5-8.6)	1.5 (0.3-5.2)	1.7 (0.3-8.6)	0.071*
Glucose (mmol/L), median (min-max)	5.0 (4.2-13.6)	5.3 (4.2-9.2)	5.1 (4.2-13.6)	0.501*
ALT (μkat/L), median (min-max)	0.5 (0.2-3.2)	0.4 (0.2-1.8)	0.5 (0.2-3.2)	0.717*
Number of previous HAART, median (min-max)	4.5 (2.0-8)	4 (3-9)	4 (3-9)	0.188*

* Chi square test. † Nucleoside tase. ‡ Nucleoside tase. NNRTI: Non Nucleoside Reverse Transcriptase Inhibitor. PI: Protease Inhibitor. VL: Viral Load. ART: antiretroviral. ††: Abacavir, AZ: zalcitabine.

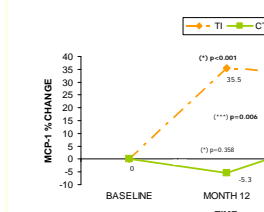
IL-8, sP-SELECTIN, t-PA, sCD40L % OF CHANGE OR LOG. CHANGE BY TREATMENT ARM



AS SHOWN IN THE GRAPH, COMPARISON BETWEEN GROUPS AT MONTH 12 AND 24 WERE NOT SIGNIFICANT. HOWEVER, DIFFERENCES INTRA GROUPS WERE DETECTED IN IL-8 AND t-PA BOTH IN MONTH 12 AND 24 (IL-8 TI group: p=0.022 and p=0.050; IL-8 CT group: p=0.011; t-PA TI group: p=0.029 and p=0.054; t-PA CT group: p=0.004 and p=0.001) AND IN sP-SELECTIN AND sCD40L IN TI GROUP AT MONTH 12 (p=0.0148 and p=0.004 respectively).

RESULTS

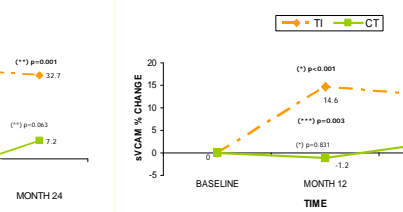
MCP-1 % OF CHANGE BY TREATMENT ARM



(*) % OF CHANGE FROM BASELINE TO MONTH 12 (Wilcoxon test)
 (***) % OF CHANGE FROM BASELINE TO MONTH 24 (Wilcoxon test)
 (***) DIFFERENCES BETWEEN ARMS AT MONTH 12 (Mann-Whitney U test)

MCP-1 MEDIAN ABSOLUTE VALUES (pg/mL)			
BASELINE	12 m	24 m	
TI	255.5	397	358
CT	317.5	314.2	371.3

sVCAM % OF CHANGE BY TREATMENT ARM



(*) % OF CHANGE FROM BASELINE TO MONTH 12 (Wilcoxon test)
 (***) % OF CHANGE FROM BASELINE TO MONTH 24 (Wilcoxon test)
 (***) DIFFERENCES BETWEEN ARMS AT MONTH 12 (Mann-Whitney U test)

sVCAM MEDIAN ABSOLUTE VALUES (ng/mL)			
BASELINE	12 m	24 m	
TI	852.5	483.8	470.5
CT	4231.7	4143.7	4107.4

CONCLUSIONS

- PATIENTS WITH VIRAL REPLICATION DUE TO HAART INTERRUPTION (DURING AT LEAST 70% OF STUDY PERIOD) PRESENTED AN INCREASE OF INFLAMMATORY MARKERS (MCP-1, sVCAM-1, IL-6) AFTER 24 MONTHS, COMPARED WITH PATIENTS UNDER CONTINUOUS THERAPY. RESULTS WERE CONFIRMED WHEN ONLY PATIENTS WHO WERE OFF TREATMENT THROUGHOUT ALL THE STUDY PERIOD WERE CONSIDERED FOR ANALYSIS
- INTERESTINGLY, THESE PROTEINS ARE RELATED WITH ENDOTHELIUM AND SUB-ENDOTHELIUM DAMAGE AT THE EARLY STEPS OF ATHEROMA PLAQUE DEVELOPMENT. THEREFORE, OUR DATA MIGHT EXPLAIN IN PART THE HARMFUL CARDIOVASCULAR EFFECT OF HAART INTERRUPTION.
- DIFFERENCES WERE NOT SO CLEAR IN THE PROTEINS KNOWN TO BE RELATED WITH PLAQUE DESTABILIZATION. ONE MAY SPECULATE THAT TO FIND ALSO DIFFERENCES IN THIS TYPE OF CYTOKINES PATIENTS SHOULD BE FOLLOWED UNTIL A CARDIOVASCULAR ACUTE EVENT.
- LONGER FOLLOW UP PERIODS AND FURTHER STUDIES ARE NEEDED TO STANDARDIZED THE USE OF CYTOKINES IN CARDIOVASCULAR RISK SCORES AND DETERMINE WHICH ARE THE PROPER BIOMARKERS FOR CARDIOVASCULAR DISEASE IN HIV-INFECTED PATIENTS.



Other members of the STOPAR* Inflammatory Markers Substudy Group:

Pochita Sanchez: Hospital Universitari de Bellvitge, Barcelona. Sergio Padilla and Enrique Bernal: Hospital General de Elche, Alicante. Montserrat Foster, Mar Gutiérrez: H. Sant Pau i Santa Creu, Barcelona. Jose Ramon Blanco: Hospital de la Rioja, Logroño.

* Clinical trial: ISRCTN75856952