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AURANOFIN PLUS NICOTINAMIDE IMPACT HIV RESERVOIR AMONG ART SUPPRESSED HIV INDIVIDUALS

Background

- Barriers for HIV persistence among individuals with suppressive antiretroviral therapy (ART) are *(i)* residual HIV replication, *(ii)* virus/cell latency, and *(iii)* viral sanctuaries.
- To measure the impact of isolated and combined strategies in decreasing viral persistence and inflammation, we investigated the effect of treatment intensification with Dolutegravir (DTG) with/without Maraviroc (MVC) in order to decrease HIV residual replication, Nicotinamide (NAM) in order to disrupt HIV latency, auranofin in order to induce lymphocyte apoptosis, especially in the memory compartment of lymphocytes, encompassing the viral reservoir, and an autologous dendritic cell vaccine to target HIV resident in latent cells and/or in sanctuaries.
- Dolutegravir was chosen due to its unprecedent potency [Min S et al, AIDS 20111]
- Maraviroc was chosen since it decreases inflammation and apoptosis [van Lelyveld SF et al, Plos One, 2015], and its capacity of latency interruption [Madrid-Elena N, J Virol 2018]
- Nicotinamide, an Histone Deacetylase (HDAC) Class II inhibitor is being explored as an anti-proliferative agent [Audrito et al. Cancer Res 2011] and an anti-HIV latency agent [Samer et al. EACS 2017].
- Gold salt auranofin decrease lymphoproliferation by inhibiting synthesis of IL-2 [Vint et al., Cell Immunol 1991] and induces lymphocyte apoptosis in the memory compartment of lymphocytes, encompassing the viral reservoir [Chirullo et al., Cell Death Dis, 2013].
- Autologous dendritic cell (DC) vaccine was used here to booster cellular immunity.

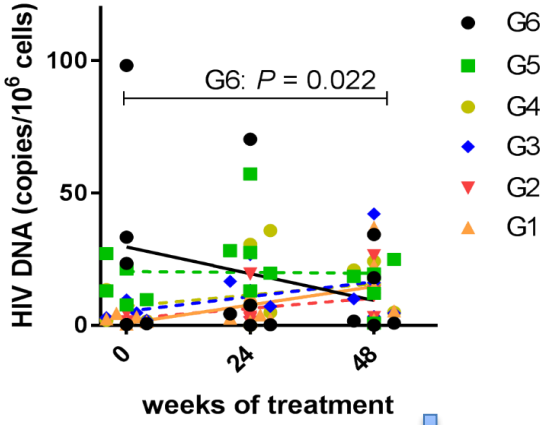
Methods:

- Study design: Randomized open label pilot proof of concept clinical trial [www.clinicaltrials.gov; ID: NCT02961829].
- Six study arms with 5 patients each followed every 4 weeks for a total of 48 weeks. Selected patients were ART suppressed for >2 years, with CD4+ T cell count nadir >350, harboring R5 HIV strains. Groups are as followed:

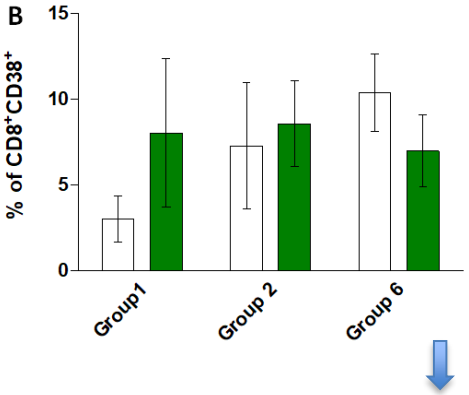
1) Continuation of ART (control group)
2) intensified ART (Continuation of ART+DTG and MVC)
3) intensified ART and HDACi (ART+DTG+MVC+NA)
4) intensified ART and Auranofin (ART+DTG+MVC+Auranofin)
5) partially intensified ART (ART+DTG), followed by DC vaccine
6) partially intensified ART (DTG)+NA+Auranofin, followed by DC vaccine.

- Auranofin was used for the first 24 weeks of the study in G4 and G6. Sera, plasma, PBMCs, saliva, urine were collected every month. Rectum biopsies were performed at baseline and at 48 weeks in groups 1, 2, 3, and 4, and at the end of DC vaccine protocol in groups 5 and 6.
- Laboratory paramethers
  - Proviral DNA quantitation in PBMCs and Rectum biopsies tissues: [Komniakis et al., Clin Microbiol 2012; Buzón et al., Nat Med 2010; Kumar et al. J Neurovirol 2007].
  - T Cell activation markers:
  - DC vaccine preparation: HIV Gag256-367 characterization followed by design of autologous GAG peptides (nanomers) according to the best immunogenicity (biding affinity > 100) based in the specific HLA profile of each individual to elicit MHC class 1 [Kai et al 2015; Benito et al 2004]. Between 2 and 6 peptides for each patient were prepared. Monocytes of each patient were obtained by cytapheresis and transformed into DCs. Exposure of DCs to autologous peptides and preparation of 3 doses of vaccine injected into axillar and inguinal subcutaneous region every 2 weeks.
  - Antibodies (Abs) quantitation: Abbott ARCHITECT HIV Ag/Ab Combo assay (Abbott, IL, USA) and LS-Vitros assay® (Ortho-Clinical Diagnostics)
- Statistical Analysis: Data were analyzed by repeated-measures ANOVA and linear regression (to show trends over time) following an appropriate transformation where necessary.

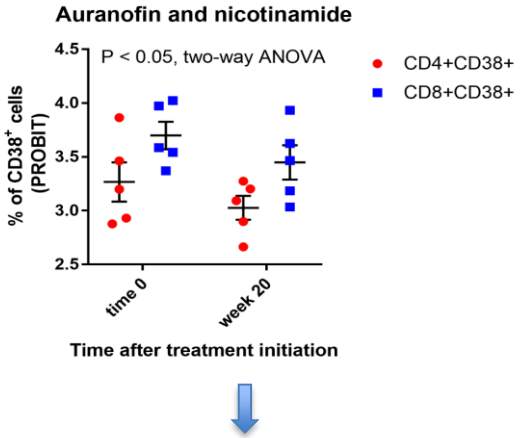
Results:



Auranofin and nicotinamide (NA) decrease proviral HIV DNA in intensified ART-treated individuals (G6) after 48 weeks of treatment, with no detected impact in the other treatment groups.



Mean % of CD8+ T cells and CD38+ at baseline (wk zero) in white bars and at week 20 in green bars among patients og G1 (control), G2 (intensification with DTG+MVC) and G6 (intensification with DTG + NA + auranofin.



Changes in the mean % of CD4+ T cells CD38+, and CD8+ T cells and CD38+ among individuals of Group 6 showing a significant decrease of these cell activation marker over time.

	wk0	wk24	wk48	2 months after the last vaccine dose
G5	9.77	27.57	12.13	124.81
	7.70	57.25	0.82	11.96
	13.00	13.00	24.92	15.63
	21.37	28.21	18.51	46.75
	27.12	19.78	19.41	31.94
Mean	**15.79**	**29.16**	**15.16**	**46.22**
G6	0.62	Bellow LOD	Bellow LOD	Bellow LOD
	33.31	4.36	34.36	63.52
	98.30	70.37	18.09	20.37
	23.43	7.65	1.65	Bellow LOD
	0.32	0.25	0.82	15.37
Mean	**31.19**	**20.66**	**13.73**	**33.09**

Levels of proviral DNA quantitation at PBMCs among patients of G5 and G6 at baseline (wk zero), week 24, week 48 and 2 months after 3 doses of autologous DC vaccine. One patient of G6 evolved to proviral DNA quantitation at PBMCs to levels below detection limits (0.001 copies10^6 cells) after treatment with intensified ART + NA + DTG, and 1 patient of G6 evolved to proviral DNA quantitation at PBMCs to levels below detection limits after DC vaccine. Two patients, 1 from G5 and other form G6, interrupted ART after week 48 and before vaccination by their own decision, and presented a rebound in the viremia, which reflected in the increase of proviral load at PBMCs (yellow mark in the Table) and an marked increase in the Antibody levels by the LS-Vitros assay ® (data not shown). No other patients from other groups interrupted ART.

Conclusion:

- Auranofin and nicotinamide (NA) significantly decrease viral DNA in intensified ART-treated individuals, with one individual evolving with undetectable levels of HIV proviral DNA in PBMCs.
- There was a decrease in the micro-inflammation levels in the auranofin and NA and ART intensification group (G6) as suggested by the activation markers in the CD4+ and CD8+ T cells.
- After Autologous Dendritic cell vaccination one individual from G6 also evolved to undetectable levels of HIV proviral DNA in PBMCs.
- Auranofin and nicotinamide administration in combination with intensified ART was well tolerated and safe.
- Results suggest that an impact of the antiproliferative approach on the viral reservoir size could be possible.
- Analytical Treatment Interruption is being planed for study participants.