

Safety and efficacy of WG-am, an innovative microbicide with dual HIV-1 inhibition derived from elite controllers

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HIV-1 remains a global health challenge, necessitating novel preventive strategies. Elite controllers (EC), a unique subset of individuals capable of naturally suppressing HIV-1 without antiretroviral therapy, are pivotal in identifying potential antiviral approaches. WG-am, a novel antiviral agent derived from EC, exhibits dual activity against HIV-1, blocking viral entry and retrotranscription.

The purpose of this study is to investigate the potential of WG-am as microbicide using human cervico-vaginal tissue (CVT) explants. Two ex-vivo tissue culture models derived from CVT from women undergoing hysterectomy. The biocompatibility and efficacy of WG-am were evaluated. FACs and multiplex analysis assessed immune cell population status and activation/inflammation. WG-am inhibited HIV-1 infection by 80% in the model. The maintenance of healthy mucosal immunity is essential for microbicide effectiveness. The biosafety of WG-am was demonstrated by studies of cytotoxicity, immune cell population, inflammation or tissue damage. No activation of T cell populations or co-receptor (CXCR4 and CCR5) deregulation were found. Further assays are being performed to fully understand the effect on WG-am on vaginal and cervical LCs using another ex-vivo tissue model.

Acknowledgement: This work has received funding from the European Union's Horizon Europe research and innovation program under grant agreement 101130633.

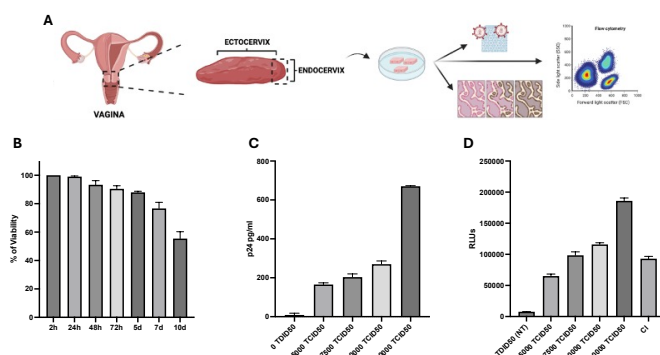


Figure 1. Tissue model and infection set up. A) Schematic representation of the human cervico-vaginal tissue model. B) Viability of the tissue up to 14 days measured by MTT assay. Tissue viability at 2h was set as 100% of viability. C) HIV-1 infection TCID50 set up. Amount of virus in the tissue SN after 72h of infection, measured by ELISA p24. D) HIV-1 infection TCID50 set up. HIV-1 infection measured by titration of T2M.b1 reporter cell line with tissue SN quantify as Relative Luminescence Units (RLUs). SN supernatant. CI is the control of infection in the second round of infection in T2M.b1, HIV-1 virus (150 TCID50) was used to directly infect the T2M.b1.

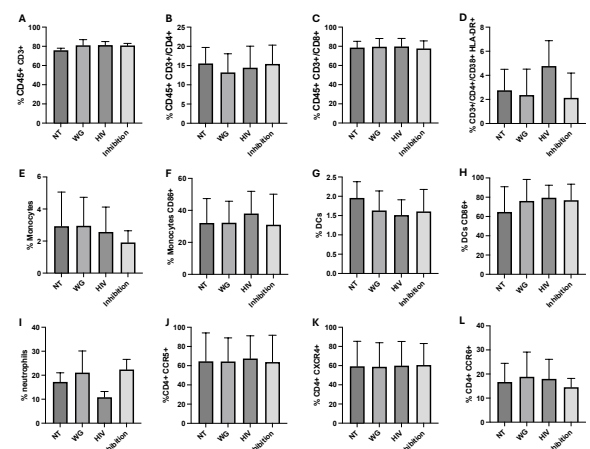


Fig. 2. Cell relative abundance in tissue explants. Effect of WG-am treatment after 72h. Cervico vaginal tissue explants were treated with WG-am 10 mM for 72h and analysed by flow cytometry. Data represent effect on WG-am on A) CD3+ B) CD3+ CD4+ C) CD3+ CD8+ D) CD4+ T Cell activation as CD38+ HLA-DR+ E) Monocytes F) Monocyte activation as CD86+ G) Dendritic cells H) Dendritic cells activation as CD86 I) Neutrophils J) CD4+ CCR5+ K) CD4+ CXCR4+ L) CD4+ CCR6+. Bars represent the mean of 4 tissue donors $\pm$ SD. NT non-treated. Inhibition WG-am 10mM + HIV-1 7500 TCID50.

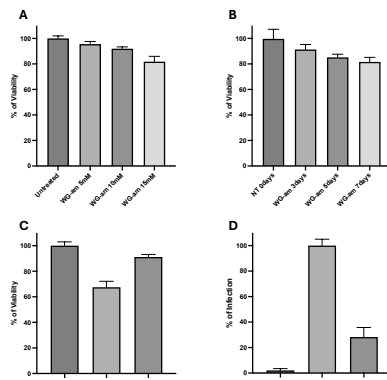


Fig. 3. Cervico-vaginal tissue viability and HIV inhibition after WG-am treatment. A) Viability of the tissue after treatment with WG-am at different concentrations (5, 10 and 15mM) for 72h. B) Viability of the tissue after treatment with 10mM WG-am after 1, 3, 5 and 7 days. C) Viability of the tissue after treatment with 10mM WG-am and infection with 7500 TCID50 HIV-1. D) HIV-1 inhibition by 10mM WG-am in cervico-vaginal tissue. Tissue viability measured by MTT assay and HIV-1 inhibition titrated on T2M.b1 reporter cell line $\pm$ SD. Data represent mean of n=3 (A, B and C) and n=6 (D) different ectocervix tissue explants made by triplicate. NT non-treated. CI control of infection.

CONCLUSIONS

- WG-am is a new antiviral compound which was found enhanced in plasma and feces of EC in comparison to VP and HC
- WG-am inhibits HIV-1 infection in ex-vivo cervicovaginal tissue model.
- WG-am is safe and does not alter immune cell populations or their activation in the ex-vivo vaginal model or in PBMCs.

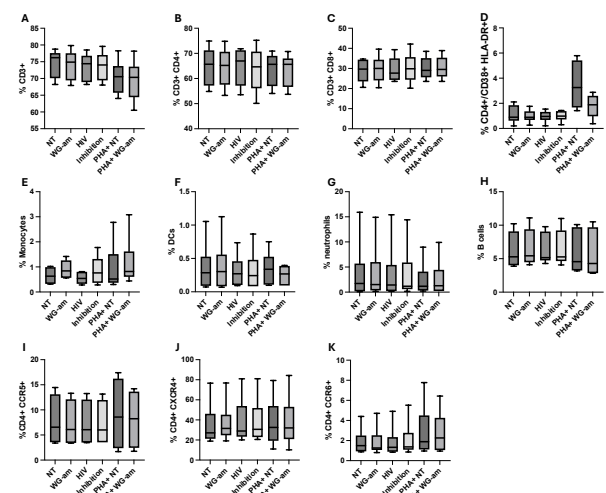


Fig. 3. Effect of WG-am treatment on PBMCs activation and cell proliferation. PBMCs isolated from buffy coats were or were not treated with PHA 2 ug/ml for 48h to induce T cell activation. Then, cells were treated with WG-am 1mM for 72h and analysed by flow cytometry. Data represent effect on WG-am on A) CD3+ B) CD3+ CD4+ C) CD3+ CD8+ D) CD4+ T Cell activation as CD38+ HLA-DR+ E) Monocytes F) Dendritic cells G) Neutrophils H) B cells I) CD4+ CCR5+ J) CD4+ CXCR4+ K) CD4+ CCR6+. Bars represent the mean of 6 buffy coat donors $\pm$ SD. NT non-treated. Inhibition WG-am 1mM + HIV-1.

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