Enhancing death of HIV-infected macrophages using BH3 mimetics.

Authors:
Brunton-O'Sullivan M[1], Rikard-Bell L[1], Kouskousis B[1], Jaworowski A[1,2], Hearps A[1,3,4].

[1] Life Sciences Discipline, Burnet Institute, Melbourne, VIC 3004, Australia

[2] Department of Infectious Diseases, School of Translational Medicine, Monash University, Melbourne, VIC 3004, Australia

[3] Department of Immunology, School of Translational Medicine, Monash University, Melbourne, VIC 3004, Australia

[4] Department of Infectious Diseases, University of Melbourne, VIC 3004, Australia

Background:
Latent HIV-infected macrophages persist in the tissues of people living with HIV despite effective treatment with antiretroviral therapy (ART) and represent a barrier to cure. Survival of HIV-infected macrophages may be associated with upregulation of anti-apoptotic proteins. BH3 mimetic drugs can enhance apoptosis by binding and inhibiting anti-apoptotic proteins and have shown promise in enhancing killing of HIV-infected T cells, however efficacy in macrophages remains unknown.

Methods:
Monocytes were isolated from healthy blood donors and differentiated into monocyte-derived macrophages in pooled human serum (MDM) and alveolar-like MDM cultured in GM-CSF (AlvMDM). Macrophages were infected with an R5-tropic HIV-GFP reporter virus and imaged after infection using the ImageXpress Pico microscope +/- BH3 mimetics: Venetoclax (BCL-2 inhibitor), S63845 (MCL-1 inhibitor) and A-1331852 (BCL-XL inhibitor).

Results:
HIV-infected MDM had enhanced viability *in vitro* when compared to mock-infected MDM (4.6% versus 7.6% non-viable cells, $p=0.044$), but an opposite trend was observed in HIV-infected AlvMDM (7.3% versus 3.9%, $p=0.096$). In uninfected macrophages, BH3 mimetics showed minimal toxicity at concentrations between 0.05-10 μM. A dose-dependent decrease in viability of HIV-infected AlvMDM was observed when treated with Venetoclax, (21.5% viability at 10 μM compared to 70% for mock,$p=0.016$). A sustained decrease in viability of HIV-infected AlvMDM compared to mock-infected cells was also seen at all concentrations of A-1331852 (all $p<0.05$). In contrast, no significant difference in viability was observed between HIV and mock-infected MDM treated with Venetoclax or A-1331852, and the MCL-1 inhibitor S63845 showed no differential effect on HIV-infected cells in either MDM or AlvMDM.

Conclusion:
HIV-infected MDM demonstrated enhanced cell survival and were resistant to BH3 mimetic-stimulated cell death. In comparison, HIV-infected AlvMDM were sensitive to BH3 mimetics targeting BCL-2 and BCL-XL. These findings highlight differences between HIV-infection in macrophage populations and provide important insights for developing strategies to eliminate HIV reservoirs.

Disclosure of Interest Statement:

None.