# Alterations in the Lymphocyte Populations in Bladders and Kidneys of Human Immunodeficiency Virus 1 Transgenic Mice

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## BACKGROUND

Urological [1] and kidney [2] complications can occur in patients living with HIV and may shorten the lifespan of patients, although current antiretroviral therapy has led to a significant improvement in the life expectancy of persons living with HIV. The spectrum of HIV associated complications can be related to the local and systemic immune response and other factors including infection and treatment.

The HIV-1 transgenic mouse model (TgFVB) have been shown to be susceptible to the HIV-1 transgene, demonstrating nephropathy [3] and other complications including neurological deficits [4], that have been associated with inflammation. However, the inflammatory cells in the renal and urological tissues of the TgFVB mice have not been fully evaluated to identify if there are alterations in the resident lymphocyte populations and the influx of inflammatory lymphocytes that maintain residency in renal and urological tissues.

In this study, we set out to evaluate the local bladder and kidney lymphocyte populations, and the splenic and circulating lymphocyte populations in the TgFVB mice.

## MATERIALS AND METHODS

- Mice were genotyped to identify WT (FVB/NJ) or HIV Transgenic Mice (TgFVB), by the presence of Env, Vpr and Vif genes
- Cells were isolated from bladders, kidneys, and spleens of WT and TgFVB
- Cells were stained with live/dead, blocked to ensure no non-specific binding, and stained with cell surface markers specific for the lymphocyte populations including CD4, CD8, and Natural Killer (NK) Cells
- The lymphocytes were analyzed using the Cytek Aurora Flow Cytometer and Flow Jo Software
- CD4+ T-cells; CD3+, CD19-, CD14-, NK1.1-, CD8- CD4+
- CD8+ T-cells; CD3+, CD19-, CD14-, NK1.1-, CD8- CD8+
- Activation markers: CD44, CD127, CD62L
- Exhaustion markers: PD-1, CTLA-4, LAG3

## RESULTS

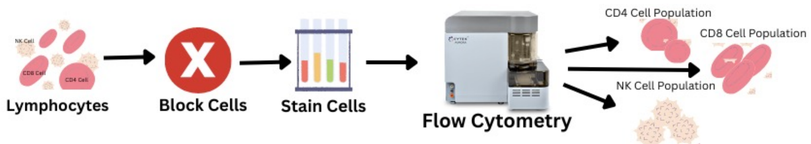


**Figure 1:** Schematic of the flow cytometry

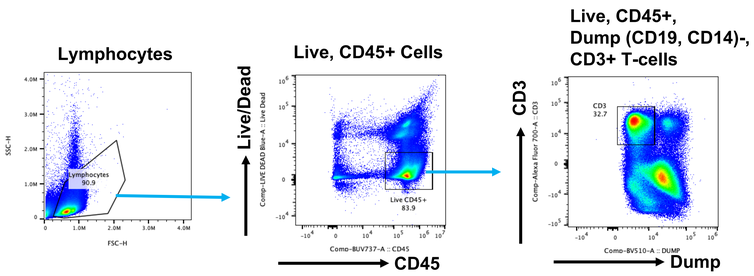


**Figure 2:** Flow cytometry gating strategy to identify the CD3+ T-cells

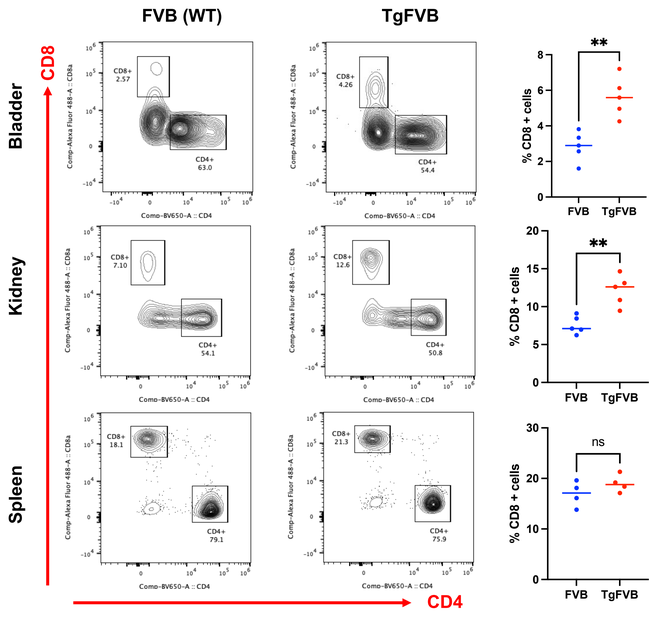


**Figure 3:** Identification of the CD4+ and CD8+ T-cell populations in the bladder (top), kidney (middle) and spleen (bottom) of FVB and TgFVB mice. Our preliminary data indicated an increase in the CD8+ T-cell population in the bladders and kidneys for the TgFVB mice in comparison to FVB mice.

## CONCLUSION

- We can detect CD4+ and CD8+ T-cells in the bladders, kidneys and spleens of TgFVB and FVB mice
- Our preliminary data indicates there are alterations in the CD8+ T-cell populations in the bladders and kidneys of TgFVB mice.
- We are currently expanding the number of TgFVB and FVB mice to evaluate the lymphocyte populations in the bladder and kidney.
- We are continuing to develop our in-depth immunophenotyping panel to characterize the inflammatory lymphocyte populations in the bladder and the kidney.

## REFERENCES

**1:** Urological aspects of HIV and AIDS. Heyns CF, et al.. Nat Rev Urol. 2013. PMID: 24166342
**2:** Kidney Diseases Associated with Human Immunodeficiency Virus Infection. Cohen SD, et al. N Engl J Med. 2018. PMID: 29694807
**3:** GWAS in Mice Maps Susceptibility to HIV-Associated Nephropathy to the Ssbp2 Locus. Steers NJ, et al. J Am Soc Nephrol. 2022. PMID: 34893534
**4:** Adult neurogenic deficits in HIV-1 Tg26 transgenic mice. Putatunda R, et al. J Neuroinflammation. 2018. PMID: 30314515

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