

## CD4 Lymphocyte Enumeration in Patients with Human Immunodeficiency Virus Infection Using Three-Colour and Four-Colour Dual-Platform Flow Cytometry: An Inter-Laboratory Comparative Evaluation

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### Abstract

**Introduction:** This study aims to evaluate the correlation and agreement between 2 methodologies of CD4 lymphocyte enumeration. **Materials and Methods:** Fifty-two blood samples from patients with human immunodeficiency virus (HIV) infection were sent for CD4 lymphocyte enumeration at 2 major hospitals using dual-platform flow cytometry where the absolute lymphocyte counts were determined on separate haematology analysers. CD4 cell enumeration was accomplished using 3-colour flow cytometry on the FACSCalibur cytometer (Becton Dickinson) in Hospital A and 4-colour flow cytometry on the Coulter Epic XL cytometer (Beckman Coulter) in Hospital B. The percentages and absolute counts of CD4 lymphocytes obtained were analysed using both linear regression and Bland-Altman plots. **Results:** On linear regression plots, the total white cell counts, absolute lymphocyte counts, CD4 lymphocyte percentages and absolute CD4 counts from the 2 hospitals correlated well with correlation coefficients,  $r > 0.95$ . On the Bland-Altman plots, there was a mean difference of  $0.13 \times 10^9/L$  and  $0.06 \times 10^9/L$  in the total white cell and absolute lymphocyte counts from the 2 hospitals respectively. The CD4 lymphocyte percentages revealed a mean difference of only 0.05% (95% limits of agreement, -3.6 to 3.7%) but there was a mean difference of 14/uL in the absolute CD4 lymphocyte counts (95% limits of agreement, -113 to 142/uL). **Conclusions:** The CD4 lymphocyte percentages obtained using the 3-colour and 4-colour flow cytometry correlated and agreed well. However, the absolute CD4 lymphocyte counts obtained using the dual-platform technique in both the hospitals did not agree well. Hence, absolute CD4 lymphocyte counts from the 2 hospitals cannot be used interchangeably in clinical practice due to poor inter-laboratory comparability.

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**Key words:** Agreement, Correlation, Dual-platform flow cytometry, Haematology analysers

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