

ABSTRACT

Aims: Sequel to the resurgence of TB co-infection in HIV/AIDS patients in sub-Saharan Africa, efavirenz has become an important component of the highly active antiretroviral treatment (HAART). The objective of this study therefore is to provide a simple reversed-phase high performance liquid chromatographic (HPLC) method for the determination of efavirenz in human plasma. Study Design: Method development and experimental study. Place and Duration of Study: School of Pharmacy, University of Nairobi, Nairobi, Kenya, between October 2009 and September 2010. Methodology: A 500 μ l drug-free plasma sample was each placed in six different centrifuge tubes (2ml) and varying aliquots of the stock solution (100 μ g/ml) of efavirenz were spiked and vortexed for 60sec to give concentrations of 0.5, 1.0, 2.0, 4.0, 8.0 and 16 μ g/ml for calibration standards and 2.0, 4.0, 8.0 and 16.0 μ g/ml for quality control samples. The off-column sample pretreatment was carried out by protein precipitation using ice-cold acetonitrile. The samples were chromatographed in a phenomenex (C18) 5 μ m particle size column with 250x4.6mm I.D and UV detection at 254nm using a mobile phase, which was made up of a mixture of solutions A and B. Both consisted of acetonitrile, 25mM ammonium acetate buffer and glacial acetic acid in proportions of 90:10:0.1 and 10:90:0.1(v/v), respectively. The analytical technique was validated for precision, accuracy and analyte recovery. Results: The calibration plot for efavirenz was found to be linear over the concentration range of 0.5 to 16.0 μ g/ml with the regression line equation obtained as $y=26842x-409.4$ and the regression coefficient ($R^2=0.999$), which allows for accurate reading of the concentrations of the test samples. The RSD (%) in intraday and interday assays ranged from 0.44 to 0.78%. Accuracy ranged from 92 to 110% and the recovery was >97%. Conclusion: This new HPLC method is simple, reproducible and cost-effective and can be used for therapeutic drug monitoring of efavirenz in HIV/AIDS patients on HAART as demonstrated in this study