Notes on the Design of Bioequivalence Study: Emtricitabine/Tenofovir Alafenamide/Dolutegravir

Notes on the design of bioequivalence studies with products invited for submission to the WHO Prequalification Unit – Medicines Assessment Team (PQT/MED) are issued to aid manufacturers with the development of their product dossier. Deviations from the approach suggested below may be considered acceptable if justified by sound scientific evidence.

The current notes should be read and followed in line with the general guidelines on submission of documentation for WHO prequalification. In particular, please consult the "Multisource (generic) pharmaceutical products: guidelines on registration requirements to establish interchangeability" in: *Fifty-seventh report of the WHO Expert Committee on Specifications for Pharmaceutical Preparations,* Geneva, World Health Organization, 2024. WHO Technical Report Series, No. 1052, Annex 8.

Below, additional specific guidance is provided on the invited immediate release products containing emtricitabine, tenofovir alafenamide (TAF), and dolutegravir.

Pharmacokinetics of emtricitabine, tenofovir alafenamide, and dolutegravir

Maximum emtricitabine plasma concentrations are observed within 0.5 to 3.0 hours of dosing in the fed state. TAF peak plasma concentrations are observed after about 1 hour after dosing in fed state. Dolutegravir T_{max} is observed at 2 to 3 hours post dose.

The elimination half-lives of emtricitabine, TAF, and dolutegravir are 10, 0.51, and 14 hours, respectively, following single doses.

Administration of emtricitabine with a high-fat meal does not affect systemic exposure ($AUC_{0\text{-}inf}$) of emtricitabine; therefore, emtricitabine may be administered with or without food.

Administration of TAF with a high-fat meal increased the systemic exposure of tenofovir alafenamide by 65%; therefore, TAF should be administered with food. However, the emtricitabine/TAF FDC comparator product is labeled to be taken with or without food.

Food increased the extent and slowed the rate of absorption of dolutegravir. The bioavailability of dolutegravir depends on meal content: low-, moderate-, and high-fat meals increased dolutegravir $AUC_{0\text{-}inf}$ by 33%, 41%, and 66%, C_{max} by 46%, 52%, and 67%, and prolonged T_{max} to 3, 4, and 5 hours, respectively, from 2 hours under fasted conditions. These increases may be clinically relevant in the presence of certain integrase class resistance. Therefore, dolutegravir is recommended to be taken with food by patients infected with HIV with integrase class resistance. Otherwise, dolutegravir can be taken with or without food.

Guidance for the design of bioequivalence studies

Taking into account the pharmacokinetic properties of emtricitabine, TAF, and dolutegravir, the following guidance with regard to the study design should be taken into account:

Design: A single-dose cross-over study is recommended.

Dose: A single oral dose of one tablet of emtricitabine/tenofovir alafenamide/dolutegravir 200/25/50 mg should be feasible. The proposed product should be compared with a combination of Descovy (Gilead Scienes Int. Inc.) and Tivicay (ViiV Healthcare Co.) in the bioequivalence study.

Fasted/fed: The bioequivalence study should be conducted in the fasted state.

Subjects: Healthy adult subjects should be recruited. It is not necessary to include patients in the bioequivalence study.

Parent or metabolite data for assessment of bioequivalence: The parent drug is considered to best reflect the biopharmaceutical quality of the product. The data for the parent compound should be used to assess bioequivalence of emtricitabine, TAF, and dolutegravir.

Sample size: Information currently available to PQT/MED indicates that the intra-subject variability for emtricitabine and dolutegravir is around 20-25%, while the intra-subject variability for TAF C_{max} is reported to be approximately 36%. These data may facilitate the calculation of a sufficient sample size for the bioequivalence study.

Washout: Taking into account the elimination half-life of emtricitabine (10 hours), TAF (0.51 hours), and dolutegravir (14 hours) in healthy volunteers, a washout period of seven days is considered sufficient to prevent carry-over.

Blood sampling: The blood sampling should be intensive for the first three hours after administration to properly characterize the C_{max} of emtricitabine and TAF and during the first six hours after administration to properly characterize the C_{max} of dolutegravir. It is not necessary to take blood samples beyond 72 hours. For example, blood samples might be taken at pre-dose, 0,083, 0.17, 0.25, 0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00, 2.25, 2.50, 2.75, 3.00, 3.50, 4.00, 4.50, 5.00, 5.50, 6.00, 6.50, 7.00, 8.00, 10.00, 12.00, 24.00, 48.00, and 72.00 h after drug administration.

Analytical considerations: Information currently available to PQT/MED indicates that it is possible to measure emtricitabine, tenofovir alafenamide and dolutegravir in human plasma using LC-MS/MS analytical methodology. The bioanalytical method should be sufficiently sensitive to detect concentrations that are 5% of the C_{max} in most profiles of each formulation (test or comparator). The bioanalytical method for each analyte should be validated in the presence of the other two analytes (see ICH Harmonised Guideline M10 for more information).

Statistical considerations: The data for emtricitabine, TAF, and dolutegravir should meet the following bioequivalence standards in a single-dose crossover design study:

- The 90% confidence interval of the relative mean AUC_{0-t} of the test to reference product should be within 80.00–125.00%
- The 90% confidence interval of the relative mean C_{max} of the test to reference product should be within 80.00–125.00%.