Notes on the Design of Bioequivalence Study: Darunavir

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 can be considered acceptable if justified by sound scientific evidence.

The current notes should be read and followed in line with the general guidelines of submission of documentation for PQ. 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 darunavir.

Pharmacokinetics of darunavir

Darunavir, co-administered with 100 mg ritonavir twice daily, is absorbed following oral administration with a T_{max} of approximately 2.5–4 hours. The absolute oral bioavailability of a single 600 mg dose of darunavir alone and after co-administration with 100 mg ritonavir twice daily is 37% and 82%, respectively.

When administered with food, the C_{max} and AUC of darunavir, co-administered with ritonavir, is approximately 30% higher relative to the fasting state. Therefore, darunavir, co-administered with ritonavir, should always be taken with food. Within the range of meals studied, darunavir exposure is similar. The total caloric content of the various meals evaluated ranged from 240 Kcal (12 g fat) to 928 Kcal (56 g fat).

The terminal elimination half-life of darunavir is approximately 15 hours when co-administered with ritonavir.

In adults, linear pharmacokinetics are observed after single dose administration over the 300–1200 mg darunavir dose range with 100 mg ritonavir.

Guidance for the design of bioequivalence studies

Taking into account the pharmacokinetic properties of darunavir, the following guidance with regard to the study design should be taken into account:

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

Dose: Due to the linear / dose-proportional pharmacokinetics of darunavir and its low solubility, the highest strength in the application to PQ (i.e., 800 mg for the 400 mg, 600 mg and 800 mg strengths) should be employed in the bioequivalence study. The study may be conducted with or without concomitant administration of ritonavir. If the study is carried out with concomitant ritonavir administration, the ritonavir dosing should be started at least two days before the administration of darunavir and maintained until the end of pharmacokinetic sampling of each treatment. A single oral dose of each formulation (test or comparator) in each study period should be employed.

Fasted/fed: The bioequivalence study should be conducted in the fed state as darunavir is recommended to be taken with food because of the increased bioavailability, irrespective of the total caloric contents of the meal. Therefore, we recommend administration with a standard breakfast, not a high-fat, high-calorie meal, as a standard breakfast is considered to be closest to real-life conditions in patients.

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 rate and extent of absorption for darunavir. The data for the parent compound should be used to assess bioequivalence of darunavir.

Sample size: Information on darunavir currently available to the PQT/MED indicates that the intra-subject variability for darunavir is around 20%. These data will facilitate the calculation of sufficient power for the single-dose cross-over bioequivalence study.

Washout: Taking into account the elimination half-life of darunavir in healthy volunteers (about 15 h), a washout period of seven days is considered sufficient to prevent carry over.

Blood sampling: The blood sampling for darunavir should be intensive the first four hours after administration to cover the rate and extend of absorption of darunavir. For example, samples should be taken at pre-dose, 0.33, 0.67, 1.00, 1.33, 1.67, 2.00, 2.33, 2.67, 3.00, 3.33, 3.67, 4.00, 4.50, 5.00, 6.00, 8.00, 10.00, 12.00, 16.00, 24.00, 36.00, 48.00 and 72.00 h after drug administration. It is not necessary to take blood samples beyond 72 hours, as this will only characterize the elimination phase darunavir.

Analytical considerations: Information currently available to the PQT/MED indicates that it is possible to measure darunavir 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 should be validated in the presence of ritonavir if the study is conducted with concomitant administration of ritonavir.

Statistical considerations: The data for darunavir 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%.