Notes on the Design of Bioequivalence Study: Clofazimine

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 for 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 clofazimine.

Pharmacokinetics of clofazimine

Following oral administration, clofazimine has a variable absorption ranging from 45% to 62% with 9% to 74% of an administered dose appearing in faeces. About 20% of a dose is absorbed from the gastrointestinal tract when clofazimine is administered as coarse crystals, but 45 to 70% of a dose may be absorbed when the drug is administered as a micronized suspension in an oil-wax base. Simultaneous ingestion of food increases the bioavailability in terms of area under the curve by 60% and tends to increase the rate of absorption. After single oral intake in the form of capsule clofazimine, unchanged plasma peak is reached in 6 to 12 hours.

Clofazimine is excreted principally in faeces, both as unabsorbed drug and via biliary elimination. Faecal elimination of clofazimine exhibits considerable interindividual variation, and 35% to 74% of a single oral dose may be excreted unchanged in faeces over the first 72 hours after the dose. Clofazimine is retained in the human body for a long time. The elimination of clofazimine is slow. In healthy subjects after single administration of 200 mg, clofazimine mean plasma elimination half-life has been reported to be 10.6 (± 4.0) days, but it has also been reported to be as little as 70 hours. The half-life following repeated oral doses is estimated to be at least 25 days.

Guidance for the design of bioequivalence studies

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

Design: In spite of the long half-life of clofazimine, a cross-over design is recommended. However, a parallel design is also acceptable.

Dose: As clofazimine is marketed as 50 mg and 100 mg tablets, a 100 mg dose using the highest tablet strength under development, should be used in the bioequivalence study since the pharmacokinetics are not reported to be non-linear.

Fasted/fed: The bioequivalence study should be conducted in the fed state as clofazimine may exhibit a higher absorption in the presence of food and it is recommended that clofazimine be taken with a meal.

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 clofazimine.

Sample size: Clofazimine AUC_{0-t} and C_{max} in the fed state have a moderate intra-subject variability (<30%). These data may facilitate the calculation of a sufficient sample size for a cross-over bioequivalence study. However, in case of a parallel design, inter-subject variability must be taken into account and these data are not presently available.

Washout: In case of a cross-over design the long-half-life of clofazimine must be taken into account, but it is not possible to define since its half-life may vary from 70 hours up to at least 10 days. Therefore, a washout period of at least 2 months i is considered sufficient to prevent carry over.

Blood sampling: The blood sampling should take into account that clofazimine absorption is slow and that T_{max} occurs after 6–12 hours. Entero-hepatic recycling seems to occur as well. Therefore, the blood sampling does not need to be very intense during the first hours, but sufficiently frequent (e.g. every 30 minutes) during the first 12 hours after administration, to properly characterize the C_{max} of clofazimine. Considering the elimination half-life, it is sufficient to take blood samples up to 72 hours after administration for the characterization of clofazimine pharmacokinetics.

Analytical considerations: Information currently available to PQT/MED indicates that it is possible to measure clofazimine 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).

Statistical considerations: The data for clofazimine should meet the following bioequivalence standards in a single-dose crossover or parallel design study:

- The 90% confidence interval of the relative mean $AUC_{0-72\,h}$ 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%.