

Notes on the design of bioequivalence study: Artesunate/Mefloquine

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 WHO prequalification. In particular, please consult the "Multisource (generic) pharmaceutical products: guidelines on registration requirements to establish interchangeability" in *Fifty-first report of the WHO Expert Committee on Specifications for Pharmaceutical Preparations*, Geneva, World Health Organization, 2017. WHO Technical Report Series, No. 1003, Annex 6.

Below, additional specific guidance is provided on the invited immediate release products containing artesunate and mefloquine.

Pharmacokinetics of artesunate and mefloquine

Artesunate is rapidly absorbed after oral administration. Most of the artesunate is promptly converted, mainly through plasma esterases, into the active metabolite dihydroartemisinin (DHA). The median (range) artesunate T_{max} value is 0.25 hours (0.25-1.33 h).

Artesunate has a plasma elimination half-life of 3–29 minutes.

When artesunate was taken with a high fat meal in healthy volunteers, the C_{max} and AUC_{0-t} of artesunate decreased 66 and 13%, respectively, compared to that observed under fasting conditions. The C_{max} and AUC_{0-t} of the active metabolite dihydroartemisinin (DHA) decreased 48 and 5%, respectively, with a high-fat meal, compared to that observed under fasting conditions. Artesunate should not be taken with a high-fat, high-calorie meal.

The maximum plasma concentration of mefloquine is reached within 6 to 24 hours after a single oral dose. The presence of food significantly enhances the rate and extent of absorption. The RBC concentration of mefloquine is almost twice as high as the plasma level.

The average elimination half-life of mefloquine in Caucasians is 21 days. Pharmacokinetic differences have been observed between various ethnic populations.

The tablets should be swallowed whole preferably after a meal with plenty of liquid.

Guidance for the design of bioequivalence studies:

Taking into account the pharmacokinetic properties of artesunate and mefloquine the following guidance with regard to the study design should be taken into account:

Design: A parallel design is recommended due to the long half-life of mefloquine. A crossover design would require a washout period of at least 100 days in principle.

Dose: As the EoI includes artesunate / mefloquine 100 / 200 mg tablets and 25 / 50 mg (preferably dispersible) tablets, the highest strength of the applied strengths should be tested. As the comparator product of mefloquine contains 250 mg and the comparator product of artesunate contains 50 mg, the results obtained for mefloquine need to be dose normalized to correct for the difference in administered dose, and two tablets of the comparator

product for artesunate need to be administered, i.e., 1 x 100/ 200 mg test product vs. 2 x 50 mg artesunate comparator + 1 x 250 mg mefloquine comparator.

When conducting bioequivalence studies, it is essential to administer the test and the comparator product according to their corresponding instructions for use. In those cases where the test and the comparator product are different dosage forms with different methods of administration (e.g., a tablet that should be taken with a glass of water vs. a dispersible tablet to be dispersed in 10 mL of water) the bioequivalence study should be conducted employing the intended method of administration of each product. It is considered incorrect to standardise the volume of liquid in all these cases (e.g. administering a glass of water after the intake of a dispersible tablet or rinsing the container where a dispersible tablet has been suspended with the remaining liquid of a glass of water) because this standardisation does not occur in real life conditions. The total volume of water employed during administration of a pediatric dispersible tablet should not exceed 50 mL.

Fasted/fed: The bioequivalence study should be conducted in the fed state with standard fat content (500-600 Kcal). A high-fat, high-calorie meal should not be employed.

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 drugs are considered to best reflect the biopharmaceutical quality of the product. The data for the parent compounds should be used to assess bioequivalence of artesunate and mefloquine.

Sample size: Limited information is available to PQT/MED on mefloquine variability. Mefloquine values for inter-subject variability seem to range between 35 to 44%. These data may facilitate the calculation of a sufficient sample size for the parallel bioequivalence study. However, a pilot study is recommended to confirm the inter-subject variability, to explore the optimal sampling times and to explore the feasibility of a cross-over design by investigating the intra-subject variability and the necessary washout for a cross-over design.

Artesunate C_{max} may exhibit high intra-subject variability (47%). Inter-subject variability in a parallel design is expected to be higher. A pilot study is recommended to estimate it.

Washout: Not applicable in case of a parallel design and, in principle, at least 100 days in case of a cross-over design. However, a shorter wash-out period may be feasible in the case of multiphasic elimination with most of the being drug eliminated in the initial phases of elimination and plasma levels below 5% of C_{max} in the final phases of elimination. This can be investigated in the pilot study recommended above.

Blood sampling: The blood sampling should be intensive around the first hour for artesunate and around 5.5 hours for mefloquine. It is not necessary to take samples after 72 hours. For example, blood samples might be taken at pre-dose and at 00.17, 00.25, 00.33, 00.50, 00.67, 00.83, 01.00, 01.25, 01.50, 01.75, 02.00, 02.50, 03.00, 03.50, 04.00, 05.00, 06.00, 07.00, 08.00, 10.00, 12.00, 24.00, 48.00, and 72.00 h after drug administration.

Analytical considerations: Information currently available PQT/MED indicates that it is possible to measure artesunate and mefloquine 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 of artesunate should be validated in the presence of mefloquine and vice versa.

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

- The 90% confidence interval of the relative mean AUC_{0-t} for artesunate and AUC_{0-72h} for mefloquine 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%.