Notes on the Design of Bioequivalence Study: Naloxone (Parenteral)

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-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 parenteral products containing naloxone.

The EoI includes intravenous, intramuscular or subcutaneous solutions for injections in vials or prefilled syringes that can be waived when the applied product is to be administered parenterally as an aqueous solution containing the same API in the same molar concentration as the comparator product and the same or similar excipients in comparable concentrations to those in the comparator product. If these conditions are not met an in vivo bioequivalence study is required.

Pharmacokinetics of naloxone

After IV administration, serum half-life in adults ranged from 30 to 81 minutes (mean 64 ± 12 minutes).

After administered to the anterolateral aspect of the thigh, the median t_{max} was 15.6 minutes (range 5.4 to 40.2 min). The mean plasma half-life of naloxone in healthy adults was 1.46 hours (14.1%CV) following a single administration.

Guidance for the design of bioequivalence studies

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

Design: A single-dose, crossover design is recommended in the intended administration route. If several routes of administration are intended, a study after IM or SC administration would suffice.

Dose: The highest strength of the series should be tested if the conditions for an additional strength biowaiver are fulfilled.

Fasted/fed: N/A

Subjects: Healthy adult subjects should be used. 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. Therefore, bioequivalence should be based on the determination of naloxone.

Sample size: No information on naloxone intra-subject variability after parenteral administration is available currently to the PQT/MED.

Washout: Taking into account the elimination half-life of naloxone in healthy volunteers(< 2 h), a washout period of 1 day is considered sufficient to prevent carry over.

Blood sampling: As naloxone is absorbed rapidly and has a short half-life, blood sampling should be intensive in the first minutes after administration to cover the peak of naloxone, e.g., pre-dose sample and samples at 2, 4, 6, 8, 10, 15, 20, 25, 30, 35, 40, 45, 60 minutes and 1.5, 2, 3, 4, 6 and 8 hours after administration.

Analytical method: Information currently available to the PQT/MED indicates that it is possible to measure naloxone in human plasma using LC-MS/MS analytical methodology. The bioanalytical method should be sufficiently sensitive to detect concentrations that are 5% of the Cmax in most profiles of each formulation (test or comparator). See Guideline on bioanalytical method validation and study sample analysis. In: WHO Technical Report Series, No. 1060, Annex 6, or the ICH Harmonised Guideline M10 for more information on bioanalytical recommendations.

Statistical considerations: The data for naloxone should meet the following bioequivalence standards in a single dose cross-over design study:

- The 90% confidence interval of the relative mean AUC_{0-t} of the test to reference product should be within 80-125%.
- The 90% confidence interval of the relative mean C_{max} of the test to reference product should be within 80-125%.
- The 90% non-parametric confidence interval of t_{max} of the test and reference product expressed as difference in minutes should be reported as supportive information to assess the equivalent onset of naloxone effect.