

Notes on the Design of Bioequivalence Study: Amoxicillin

Notes on the design of bioequivalence studies with products invited for submission to the WHO Prequalification Team - Medicines (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 products containing amoxicillin.

Pharmacokinetics of amoxicillin

Amoxicillin is stable in the gastric acid secretion and is rapidly absorbed from the gastrointestinal tract after oral administration. The time to peak plasma concentration (T_{max}) is approximately one hour.

In the dose range 250 to 3000 mg the bioavailability is linear in proportion to dose (measured as C_{max} and AUC). Absorption is not influenced by simultaneous food intake.

Amoxicillin has a mean elimination half-life of approximately one hour.

Guidance for the design of bioequivalence studies:

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

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

Dose: As the EoI includes two strengths, 125 and 250 mg (scored) dispersible tablets, the highest strength should be used in the bioequivalence study if the conditions for a biowaiver for the low strength are fulfilled. Otherwise both strengths should be tested.

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

For administration, the test product should be dispersed in a small amount of liquid suitable for the intended paediatric population (e.g. 20 – 40 ml) and a similar small amount of water should be used to rinse the container. Additional water should not be administered in order to mimic the real conditions of use. The method of administration should be consistent with the proposed labelling for the product.

The reference product should be administered in a fashion consistent with its labelling, using a volume of water appropriate for a pediatric population. For example, for Amoxil/Clamoxyl powder for oral suspension in sachet it is indicated that the content of the sachet be suspended in 10 – 20 ml of water and administered. It is acceptable to rinse the container with an additional similar volume of water but, additional water beyond that 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 drug is considered to best reflect the biopharmaceutical quality of the product. The data for the parent compound should be used to assess bioequivalence.

Sample size: Based on the information available to PQT/MED, amoxicillin C_{max} exhibits low to moderate intra-subject variability (10-20%), whereas AUC_{0-t} exhibits low variability (10-13%) in the fasted state. These data may facilitate the calculation of a sufficient sample size for a single-dose cross-over bioequivalence study.

Washout: Taking into account the elimination half-life of amoxicillin in healthy volunteer (1 h), a washout period of three (3) days is considered sufficient to prevent carry over.

Blood sampling: The blood sampling for amoxicillin should be intensive the first four hours after administration to properly characterize the C_{max} of amoxicillin. It is not necessary to take blood samples beyond 12 hours for the characterization of amoxicillin pharmacokinetics. Samples may be taken at the following time points: Pre-dose, 0.25, 0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00, 2.50, 3.00, 3.50, 4.00, 5.00, 6.00, 7.00, 8.00, 10.00, and 12.00 h after drug administration.

Analytical considerations: Information currently available to PQT/MED indicates that it is possible to measure amoxicillin 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 amoxicillin 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 80.00–125.00%
- The 90% confidence interval of the relative mean C_{max} of the test to reference product should be within 80.00–125.00%.