Notes on the Design of Bioequivalence Study:

Clarithromycin / Rifampicin

Notes on the design of bioequivalence studies with products invited for submission to the WHO Prequalification Team: medicines (PQTm) 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.


Below, additional specific guidance is provided on the invited fixed dose combination products, containing clarithromycin and rifampicin 250/300 mg scored tablets.

Pharmacokinetics of clarithromycin

Clarithromycin is rapidly and well absorbed from the gastro-intestinal tract after oral administration. However, it exhibits an extensive first pass effect metabolism. Absolute bioavailability after the administration of a 250 mg tablet is 50% approximately. The microbiologically active metabolite 14-hydroxyclarithromycin is formed by first pass metabolism. Clarithromycin may be given without regard to meals as food does not affect the extent of bioavailability of clarithromycin tablets. Food does slightly delay the onset of absorption of Clarithromycin and formation of the 14-hydroxymetabolite. Peak serum concentrations occur about 1 to 2 hours after a single dose on an empty stomach.

The pharmacokinetics of Clarithromycin are non-linear due to saturation of the hepatic metabolism at high doses. Half-life increases from 2-4 h after the administration of 250 mg b.i.d. to 5 h after the administration of 500 mg b.i.d. The half-life of the active metabolite is 5-6 h after 250 mg doses b.i.d. At 250 mg b.i.d. 15-20% of unchanged drug is excreted in the urine. With 500 mg b.i.d. daily dosing urinary excretion is greater (approximately 36%). The 14-hydroxyclarithromycin is the major urinary metabolite and accounts for 10-15% of the dose. Most of the remainder of the dose (70-80%) is eliminated in the faeces, primarily via the bile. A small fraction (5-10%) of the parent drug is recovered from the faeces.

Pharmacokinetics of rifampicin

Rifampicin is readily absorbed from the gastrointestinal tract. Peak serum concentrations occur about 2 to 4 hours after a single dose on an empty stomach. Absorption of rifampicin is reduced when the drug is ingested with food. In normal subjects the half-life of rifampicin in serum averages about 3 hours after a 600 mg dose and increases to 5.1 hours after a 900 mg dose.

Guidance for the design of bioequivalence studies:

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

Study design: A cross-over design is recommended.
**Dose:** As the EoI includes the strength combination 250/300 mg, this combination strength should be administered versus the mono-component comparators: 250 mg of clarithromycin and 300 mg of rifampicin, administered simultaneously.

**Fasting/fed:** The bioequivalence study should be conducted in the fasting state.

**Subjects:** Healthy adult subjects should be recruited. It is not necessary to include patients in the bioequivalence study.

**Sample size:** Clarithromycin $C_{\text{max}}$ exhibits moderate or high intra-subject variability (27-35%), whereas $\text{AUC}_{0-t}$ exhibits low variability (12-19%) in the fasting state. Rifampicin pharmacokinetic parameters, $C_{\text{max}}$ and $\text{AUC}_{0-t}$, in the fasting state seem to possess low to moderate variability (10-27%), based on information available to the PQTm. These data will facilitate the calculation of a sufficient sample size for the bioequivalence study.

**Washout:** 7 days.

**Blood sampling:** Predose, 0.25, 0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00, 2.25, 2.50, 2.75, 3.00, 3.25, 3.50, 3.75, 4.00, 4.50, 5.00, 6.00, 8.00, 10.00, 12.00, 16.00, 20.00, and 24.00 h after drug administration.

**Analytical method:** Information currently available indicates that it is possible to measure clarithromycin and rifampicin in human plasma using LC-MS/MS analytical methodology. The bioanalytical method should be sufficiently sensitive to detect concentrations that are 5% of the $C_{\text{max}}$ in most profiles of each formulation (test or comparator).

**Parent or metabolite data for assessment of bioequivalence:** The parent drug is considered to best reflect the biopharmaceutical quality of the proposed product. The data for the parent compound should be used to assess bioequivalence.

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

- The 90% confidence interval of the relative mean $\text{AUC}_{0-t}$ of the test to reference product should be within 80–125%.
- The 90% confidence interval of the relative mean $C_{\text{max}}$ of the test to reference product should be within 80–125%.

In case of a single dose, replicate crossover design study, the $C_{\text{max}}$ acceptance range can be widened based on the intra-subject variability observed for the reference product since information currently available to PQTm indicates that the comparator product might be a highly variable drug product. Therefore, if the applicant suspects that the variability of $C_{\text{max}}$ is high (CV > 30%), the applicant may prefer to employ a replicate design study for at least the comparator product in order to scale the acceptance range of $C_{\text{max}}$. For more information on replicate study designs and average scaled bioequivalence, refer to Section 7.9.3 of Annex 6, TRS 1003.