Contains Nonbinding Recommendations

Draft Guidance on Amphotericin B

This draft guidance, when finalized, will represent the current thinking of the Food and Drug Administration (FDA, or the Agency) on this topic. It does not establish any rights for any person and is not binding on FDA or the public. You can use an alternative approach if it satisfies the requirements of the applicable statutes and regulations. To discuss an alternative approach, contact the Office of Generic Drugs.

Active Ingredient: Amphotericin B

Dosage Form; Route: Liposome injection; intravenous

Recommended Studies: Two studies

When the test and reference liposome products

- have the same drug product composition and
- have equivalent liposome characteristics, including liposome composition, liposome size distribution, number of lamellar, electrical surface potential or charge, lipid bilayer phase transition, and in vitro leakage rates.

The following in vivo and in vitro studies are recommended to demonstrate bioequivalence:

In Vivo Study:

A. Type of study: In vivo steady-state
Design: (1) Parallel group or (2) two-way crossover
Strength: 50 mg/vial
Dose: Appropriate dose that is recommended by physicians to fit the patients' needs per the RLD labeling
Subjects: Patients who need Amphotericin B treatment as part of their therapy per the RLD labeling. FDA recommends that the study not be conducted using healthy subjects.
Additional Comments: Please see comments below.

1. Submission of a Bio Investigational New Drug Application (Bio-IND) is required prior to conducting a bioequivalence (BE) study for a cytotoxic drug product such as Amphotericin B (see 21CFR § 320.31).

2. Following patients should be excluded from the study:

    - Less than 18 years of age
    - Pregnant or lactating women
    - History of hypersensitivity reactions to any components of conventional or liposomal Amphotericin B formulations
    - Serum creatinine concentration greater than twice the upper limit of normal (ULN) AST or ALT value greater than 10 times the ULN

3. Following safety monitoring measures are recommended for the in vivo BE study:

    - Pregnancy test for women

- Laboratory evaluation of renal, hepatic and hematopoietic function
- Serum electrolytes (particularly magnesium and potassium) at baseline and during the course of the study

4. Each qualified patient should receive the same dose of Amphotericin B Liposomal injection treatment at fixed 24-hour intervals. Exclude patients from the study with dose adjustment during the BE study. For crossover studies, patients may receive either the generic formulation (Treatment A) or the reference formulation (Treatment B) for 5 days (Day 1-5), and then switch to the other treatment for 5 days (Day 6-10). No washout period is necessary between the two treatments.

5. The patients shall receive their own established Amphotericin B dosing regimen during the study. After the study is completed, continue the patients on their Liposomal Amphotericin B Injection dosing regimens.

6. Patient's concomitant medications should remain exactly the same during the study.

7. All study subjects can be given a standard non-high-fat diet every day during the study provided there is no interference with patients' care. Alternatively, initiate the treatment two hours after a standard (non-high-fat) breakfast every day during the study. In addition, avoid concomitant therapy with intravenous fat emulsions, such as total parental nutrition (TPN), since that may change the pharmacokinetic (PK) profile of Amphotericin B Liposomal injection.

8. Blood sampling:

   For each period, two or three trough concentrations are recommended to be measured before the full PK sampling day to ensure steady-state is reached. On the day of blood sample collection, a series of blood samples should be collected to assess the concentration-time curve.

9. Statistical analysis of PK data:

   For statistical analysis of PK data, please refer to *Bioequivalence Recommendations for Specific Products — Clozapine (final)*.

10. Clinical report and adverse reactions:

    Report patient medical histories, physical examination laboratory reports, and all incidents of possible adverse reactions.

Analytes to measure (in appropriate biological fluid): Free Amphotericin B (liposome-unbound) and liposome-encapsulated Amphotericin B in plasma

Bioequivalence based on (90% CI): Free Amphotericin B and liposome-encapsulated Amphotericin B

Note: the pivotal BE study should be conducted using test product produced by the proposed commercial scale manufacturing process.

In Vitro Study:

B. Type of study: Liposome size distribution
Design: In vitro bioequivalence study on at least three lots of both test and reference products

Equivalence based on (95% CI): Population bioequivalence based on D_{50} and SPAN [i.e. $(D_{90}-D_{10})/D_{50}$] or alternatively on the harmonic intensity-weighted average particle diameter and polydispersity index derived from cumulant analysis of the size intensity distribution.

Waiver request of in vivo testing: Not Applicable.

Dissolution test method and sampling times: The dissolution information for this drug product can be found on the FDA-Recommended Dissolution Methods Web site, available to the public at the following location: http://www.accessdata.fda.gov/scripts/cder/dissolution/. Conduct comparative dissolution testing on 12 dosage units each of all strengths of the test and reference products. Specifications will be determined upon review of the abbreviated new drug application (ANDA).

Additional information:

Same drug product composition

Per 21 CFR § 314.94 (a) (9) (iii), as a parenteral drug product, a generic Amphotericin B liposome injection must be qualitatively (Q1) and quantitatively (Q2) the same as the RLD, except differences in buffers, preservatives and antioxidants provided that the applicant identifies and characterizes these differences and demonstrates that the differences do not impact the safety/efficacy profile of the drug product. Currently, FDA has no recommendations for the type of studies that would be needed to demonstrate that differences in buffers, preservatives, and antioxidants do not affect the safety/efficacy profile of the drug product.

Lipid excipients are critical in the liposome formulation. ANDA sponsors should obtain lipids from the same category of synthesis route (natural or synthetic) as found in the RLD. Information concerning the chemistry, manufacturing, and control of the lipid components should be provided at the same level of detail expected for a drug substance as suggested in the liposome drug products draft guidance. ANDA sponsors should have specification on lipid excipients that are similar to those used to produce the RLD. Provide additional comparative characterization (beyond meeting specifications) of lipid excipients including the distribution of the molecular species.

Equivalent liposome characteristics

Additional in vitro characterization are recommended to demonstrate the sameness between the test and reference products in terms of liposome composition, liposome morphology and number of lamellar, lipid bilayer phase transitions, liposome size distribution, electrical

surface potential or charge, and in vitro leakage rates. The in vitro liposome characterization tests should be conducted on at least three batches of the ANDA and RLD products (at least one ANDA batch should be produced by the commercial scale process and used in the in vivo BE study).

For additional information regarding statistical analysis of in vitro data, please refer to *Bioequivalence Recommendations for Specific Products — Budesonide Suspension (Draft)*.