Journal of Pharmaceutical and Scientific Innovation
www.jpsionline.com
Review Article

BIOEQUIVALENCE STUDY: OVERVIEW

Balkrishana D. Tiwari[1]*, Omkar N. Shikare[2], Amruta M. Sontakke[1]
[1]Sanjay Rathod Shikshan Sanstha's Manohar Naik Institute of Pharmacy, Umarkhed Dist. Yavatmal, Maharashtra, India
[2]Padmashree Dr. D. Y. Patil Institute of Pharmaceutical Sciences and Research, Pimpri, Pune, Maharashtra, India
*Corresponding Author Email: tiwaribd@ymail.com
DOI: 10.7897/2277-4572.035187

Received on: 12/09/14 Revised on: 09/10/14 Accepted on: 16/10/14

ABSTRACT
While manufacturing generic drugs, the drug companies use the same active ingredients and, they have the same risks and benefits as their brand name likeness. Also, generic drugs have the same quality, strength, purity and stability as brand name drugs. The generic drugs are less expensive as compared to branded drugs as generic manufacturers do not have the investment costs of the developer of a new drug. New drugs are generally developed under patent protection. Bioequivalence is based on rate and extent, but rate and extent is depending on the absorption. Absorption is depending on drug dissolution, permeability and solubility. Firstly permeability and solubility related to biopharmaceutical classification system discovered by Gordon Amidon *et al*. in 1995. Bio analysis carried out with help of LC/MS and use of Solid phase extraction, Liquid/Liquid extraction, Protein precipitation.
Keywords: Bioequivalence, Bio analysis, Biopharmaceutical classification system.

INTRODUCTION

The key reason for performing bioequivalence testing is to ensure that the quality of generic drug products. In particular, such testing is meant to establish that there are not likely to be any differences in safety and efficacy between a generic and an innovator drug product (reference product); that is, that the products are therapeutically equivalent. Thus, in essence, bioequivalence is considered a foster of therapeutic equivalence[1]. While manufacturing generic drugs, the drug companies use the same active ingredients and, they have the same risks and benefits as their brand name likeness. Also, generic drugs have the same quality, strength, purity and stability as brand name drugs. It is seen that Generic Drugs work in the same way and in the same amount of time as branded drugs. The generic drugs are less expensive as compared to branded drugs as generic manufacturers do not have the investment costs of the developer of a new drug. New drugs are generally developed under patent protection. The studies should provide an objective means of critically assessing the possibility of substituting one for the other. Two products marketed by different licensees, containing same active ingredients, must be shown to be therapeutically equivalent to one another in order to be considered interchangeable. Thus, because of the importance of generic drugs in health care, it is imperative that the pharmaceutical quality, safety and efficacy of generics should be reliably compared with the corresponding innovator drugs (brand - name drugs). According to Hatch-Waxman act the expansion of the generic drug industry, before the Act only 12 % of all prescriptions in the United States were generics, and in 2007, 65 % of prescriptions dispensed in the U.S were for generic drugs (Generic Pharmaceutical Association, 2008).

Table 1: Differences between generic drug and new drug

Generic Drug	New drug
Chemistry	Chemistry
Manufacturing	Manufacturing
Controls	Controls
Labeling	Labeling
Testing	Testing
Bioequivalence	Preclinical study Clinical study Bioavailability

The US Food and Drug Administration (FDA) publish a list of drug products and equivalents, Approved Drug Products with Therapeutic Equivalence Evaluations, commonly known as the "Orange Book". The FDA's designation of "therapeutic equivalence" indicates that the generic formulation is (among other things) bioequivalent to the innovator formulation and signifies the FDA's expectation that the formulations are likely "to have equivalent clinical effect and no difference in their potential for adverse effects"[2]. The assessment of "interchangeability" between the innovator and generic products is carried out by a study of "in vivo equivalence" or "bioequivalence". The apposite situations in which bioequivalence studies are required include;

- when the proposed marketed dosage form is different from that used in pivotal clinical trials;
- when significant changes are made in the manufacture of the marketed formulation;
- When a new generic product is tested against the innovator's marketed product. Based on this background, bioavailability (BA) and bioequivalence (BE) information has been determined to have practical and public health value for pharmaceutical industries, regulatory agencies, patients and practitioners. To understand the basis of the argument around innovator drug and generic exchangeability a thorough understanding of the terms associated with generic drugs is needed.

Bioavailability

"Bioavailability means the rate and the extent to which the active drug ingredient of therapeutic moiety is absorbed from a drug product and becomes available at the site of action" (FDA Official Statement in 1977), "The rate at which, and the extent to which the drug substance and/or its active metabolites reaches the systemic circulation." (International Consensus Statement in 1991)

Bioequivalence

A relative term which denotes that the drug substance in two or more dosage forms, reaches the systemic circulation at the same relative rate and to the same relative extent i.e., their plasma concentration time profiles will be identical without

significant statistical difference. Bioequivalence is based on rate and extent, but rate and extent is depending on the absorption. Absorption is depending on drug dissolution, permeability and solubility. Firstly permeability and solubility related to biopharmaceutical classification system discovered by Gordon Amidon *et al*. in 1995. The BCS used *in vitro* dissolution to establish bioequivalence for highly soluble and highly permeable compounds[3].

Table 2: BCS classification and bioequivalence absorption

BCS Class	Description	Bioequivalence
I	High Solubility – High Permeability	Very good
II	Low Solubility – High Permeability	Good
III	High Solubility – Low Permeability	Poor
IV	Low Solubility – Low Permeability	Very poor

BCS is based on scientific framework describing three rate limiting steps in oral absorption. The three necessary steps for a drug to be absorbed are:

- Free of drug from dosage forms.
- Sustainment of dissolved state through Gastro-intestinal (G.I) tract.
- Suffusion through G.I. membrane into hepatic circulation[4].

The BCS became a tool in the regulation of bioequivalence of oral drug products. Due to substantial credit of the BA/BE concept all over the world, tremendous advancements have been made by the FDA as well as various national, international and supranational regulatory authorities. In parallel, pharmaceutical industry and academia are also contributing exclusively in the area of assessment of BE[5]. Currently available advances to determine BE of generic products are largely standardized due to discussion and consensus reached among various stakeholders at numerous national and meetings, conferences, and workshops (e.g. American Association of Pharmaceutical Scientists, Federation International Pharmaceutique). Thus the currently available scientific and regulatory guidance documents are due to the combined efforts of industry, academia and regulatory scientists. Every country now has its own individual regulatory authority as well as regulatory guidance for BA/BE studies and the magnitude of judgment of BE of drug product is influenced by the regulatory environment of the respective country of marketing. The regulatory authorities of some of the countries are listed below Table[6,7].

Table 3: Regulatory Authorities of Various Countries

Country	Regulatory Authority
India	Central Drugs Standard Control Organization
United States	US Food and Drug Administration
Europe	European Medicines Agency
Canada	Health Canada
Australia	Therapeutic Good Administration
Brazil	National Health Surveillance Agency
Japan	Ministry of Health, Labour and Welfare
New Zealand	Medicines and Medical Devices Safety Authority
China	National Institute for the Control of Pharmaceutical and Biological Products
Mexico	Federal Commission for Protection against Health Risks
Germany	Federal Institute for Drugs and Medical Devices
Greece	National Organization for Medicines
United Kingdom	National Institute for Biological Standards and Control
Spain	Ministry of Health and Consumption (in Spanish)
South Africa	Department of Health Medicines Control Council

Thus there is a greater need to harmonize the regulatory environment globally for BE assessment as far as practicable so that the drug product marketed in different parts and regions of the world would have optimum drug product quality in terms of fungibility.

Methods of Assessments

The assessment of BE of different drug products is based on the central assumption that two products are equivalent when the rate and extent of absorption of the test/generic drug does not show a significant difference from the rate and extent of absorption of the reference/brand drug under similar experimental conditions as defined. As per the different regulatory authorities, BE studies are generally classified as[8-11].

- Pharmacokinetic endpoint studies.
- Pharmacodynamic endpoint studies.
- Clinical endpoint studies.
- *In vitro* endpoint studies.

Table 4: Methods of Assessments

Classification of BE studies	Description
Pharmacokinetic endpoint studies	Drug level can be determined in an easily approachable biological fluid (such as plasma, blood, urine) and drug level is correlated with the clinical result.
Pharmacodynamic endpoint studies	In such cases, BA may be evaluated and BE may be established, based on a Pharmacodynamic study, providing an appropriate Pharmacodynamic endpoint is available. Pharmacodynamic evaluation is evaluate of the effect on a pathophysiological process, such as a function of time, after administration of two different products to do as a basis for BE assessment.
Clinical endpoint studies	In the absence of pharmacokinetic and Pharmacodynamic accesses, adequate and well-controlled clinical trials may be used to establish BA/BE. Several international regulatory authorities provide general information about the conduct of clinical studies to demonstrate BE.
***In vitro* endpoint studies**	Using this BCS accesses, a highly permeable, highly soluble drug substance formulated into a rapidly dissolving drug product may need only in vitro dissolution studies to establish BE.

Clinical Phase (Bioequivalence) study includes

- Pre-study
- During study
- Post study.

In the pre-study before trial starting the test are performed by phlebotomist and those subject normally fit that includes in trial otherwise they are omitted and in during study (Fed or Fast) after dosing the blood collected and centrifuged at 3500 RPM stored at -70°C temperature. The sample collected as Analytical sample and Control sample with proper labeling. Analytical sample are analyzed with help of LC/MS. Control sample stored at ± 3 years. In post study the subject vital sign and other test are performed[12-15].

Table 5: Pre-study, during study and Post study

Pre-Study	During Study	Post Study
Screening Inform Consent Form Registration Physical Examination ECG Sample Collection Radiologic Examination. Drug Abuse Test. Blood/Urine Test. Breath Alcohol Test	Dosing Sample Collection Isolation	Weight measurement Vital sign Lab test ECG

Bio analysis

In the bioequivalence study bio analysis carried out with help of LC/MS and use of following three processes used[16].

1. Solid phase extraction.
2. Liquid/Liquid extraction.
3. Protein precipitation.

Solid phase extraction

In solid phase extraction the analyte is deposited on stationary phase then pass buffer solution, absorption takes place and analyte dissolve these shows percentage concentrate in LC/MS.

Liquid/Liquid extraction

In Liquid/Liquid extraction all the liquid extraction is evaporated and only the analyte remain the pass mobile phase which shows concentration in LC/MS.

Protein precipitation

In Protein precipitation the plasma and buffer ammonium for mate added then precipitate protein and cooling in centrifuge for 30 minutes; then lastly decanted only upper layer spiked/aliquot which shows in LC/MS[17-20].

Labeling

HPLC vial arranged in the LC/MS are three ways by different method in (Method development basis).

11:4:29:4

4:29:4

4:29:4

11: These are LQC means Lower / Lowest quality control (Range of 40 to 50 %).

MQC means Medium quality control (Range of 55 to 70 %)

HQC means High quality control (Range of 70 to 85 %).

4: These for the quality controls.

29: These for the subject number.

11: These are the blank, without concentration and also increasing concentration.

On the above information determine the exact concentration of the unknown drug matching with known drug[21-24].

Biopharmaceutical Classification System

During the last two and half decade, bio pharmaceutics has been undergoing a revolution from drug discovery to drug regulatory standards and harmonization. Bio pharmaceutics is based on the chemical and physical properties of drug content and the formulation, physiology of the route of administration. Nowadays, many particles are classified through screening processes and promising candidates enter into drug pipelines for further *in vitro* and *in vivo* tests. At the end of the development process stands the approval by the regulatory agencies[25- 28.]

Table 6: Biopharmaceutical Classification System

Class	Properties of Class
Class I	High Solubility and High Permeability
Class II	Low Solubility and High Permeability
Class III	High Solubility and Low Permeability
Class IV	Low Solubility and Low Permeability

Table 7: Some of the example of Biopharmaceutical Classification system

Drug Name	Biopharmaceutical Classification system Class
Co-amoxiclav Tablet 1000 mg	I/III
Irbesartan Tablet 300 mg	II
Cyclobenzaprine HCL ER Capsule	I
Metoprolol Tablet 100 mg	I
Carbamazepine 400 mg Divi.Tablet	II
Atorvastatin Tablet 80 mg	II
Levothyroxine Na 300 mcg	I/III
Misoprostol 200 mcg Tablet	III
Esomeprazole Tablet 20 mg	III
Tramadol Tablet 100 mg	I
Linezolide Tablet 100 mg	IV
Gabapentine 600 mg Tablet	III
Bosentan 125 mg	II
Allopurinol Tablet 300 mg	III/I
Efavirenz Tablet 600 mg	II
Emtricitabine and Tenofovir	I and III
Ritonavir 100 mg	II/IV
Fosamprenavir Calcium Tablet 700 mg	II
Pantoprazole 40 mg Capsule	III
Omeprazole Na	II
Actazolamide ER	IV
Fenofibrate 120 mg Tablet	II
Griseofulvin 250 mg	II/IV
Diclofenac SR	II
Cefuroxime Axetile 500 mg Tablet	IV
Acitretin 100 mg Capsule	IV
Aspirin 81 mg	III
Nifedipine 60 mg ER	II
Montelukast 5 mg chewable Tablet	I
Solifenacin Tablet	III
Bicalutamide Tablet 50 mg	II
Rabeprazole	I/III
Dutasteride Capule 0.5 mg	II/IV
Rosuvastatin 40 mg	III
Calcitriol 0.5 mcg	II/IV
Lisinopril Tablet 40 mg	III
Valsartan HCTZ	II
Budesonide 3 mg	II
Cefdinir Capsule 300 mg	IV
Sertraline	II
Escitalopram	I
Perphenazine	I

The BCS system helps for drug solubility and permeability, that drug with high solubility and high permeability which cross rapidly plasma membrane and shows quick action[29-34].

CONCLUSION

In bioequivalence study the use of test drug correlating with reference drug and in this study use of ingredient are same as that of the reference drug. Thus the bioequivalence study drug stability, safety, strength are same compare to brand drug. Here general discussion of bioequivalence study.

REFERENCES

1. Roger LN, Lloyd NS. Bioequivalence Requirements for Generic Products. Pharmac Ther 1994; 62: 41-55. http://dx.doi.org/10.1016 /0163-7258(94)90004-3
2. US Food and Drug Administration. Orange Book: Approved Drug Products with Therapeutic Equivalence Evaluations; 2010.
3. Cook J, William A and Henry WY. Application of the Biopharmaceutical Classification System in Clinical Drug Development-An Industrial View. The AAPS Journal 2008; 10(2): 306-310. http://dx.doi.org/10.1208/s12248-008-9036-5
4. Ku MS. Use of the Biopharmaceutical Classification System in Early Drug Development, the AAPS Journal 2008; 10(1): 208-212.
5. Meyer MC, Straughn AB, Jarvi EJ, *et al*. The bioinequivalence of carbamazepine tablets with a history of clinical failures. Pharm. Res 1992; 9: 1612-1616. http://dx.doi.org/10.1023/A:1015872626887
6. Tamboli A, Todkar P, Zope P, Sayyad F. An Overview on Bioequivalence: Regulatory consideration for Generic Drug Products 2010; 2(10): 86-92.
7. Chien J, Friedrich S, Heathman M, Alwis D, Sinha V. Pharmacokinetics or Pharmacodynamics and the stages of Drug Development: Role of Modeling and Simulation. The AAPS Journal 2005; 7(3): 544-559. http://dx.doi.org/10.1208/aapsj070355
8. Guidance for Industry, Immediate Release Solid Oral Dosage Forms Center for Drug Evaluation and Research (CDER); 1995. p. 23-25.
9. Pandey S, Pandey P, Tiwari G, *et al*. Bio analysis in the drug discovery and development. Pharmaceutical Methods 2010; 1(1): 14-23. http:// dx.doi.org/10.4103/2229-4708.72223
10. WHO. Annex 7 Multisource (generic) pharmaceutical products: guidelines on registration requirements to establish inter changeability. WHO Technical Report Series, No. 2006; 937: 347–390.
11. FDA. Guidance for industry population pharmacokinetics; 1999. p. 1–35.
12. FDA. Guidance for industry bioavailability and bioequivalence studies for orally administered drug products – general considerations; 2003. p. 1–26.
13. HC. Guidance for industry preparation of veterinary abbreviated new drug submissions – generic drugs; 2010. p. 1–71.
14. FDA. Bioequivalence guidance; 2006. p. 1–28.
15. Kohli K, Chopra S, Dhar D, Arora S, Khar RK. Self-emulsifying drug delivery systems: an approach to enhance oral bioavailability. Drug Discovery Today 2010; 15: 958–965. http://dx.doi.org/10.1016/j.drudis. 2010.08.007
16. Mueller EA, Kovarik JM, Van Bree JB, Tetzloff W, Grevel J, Kutz K. Improved dose linearity of cyclosporine pharmacokinetics from a microemulsion formulation. Pharmaceutical Research 1994; 11: 301–304. http://dx.doi.org/10.1023/A:1018923912135
17. Chiou WL, Riegelman S. Pharmaceutical applications of solid dispersion systems. Journal of Pharmaceutical Sciences 1971; 60: 1281–1302. http://dx.doi.org/10.1002/jps.2600600902
18. Vasconcelos T, Sarmento B, Costa P. Solid dispersions as strategy to improve oral bioavailability of poor water soluble drugs. Drug Discovery Today 2007; 12: 1068–1075. http://dx.doi.org/10.1016/ j.drudis.2007.09.005
19. Guidelines for ICH Harmonized Tripartite Good Clinical Practice E6; 1996. p. 8-52.
20. Crowe BJ, Xia HA, Berlin JA, *et al*. Recommendations for safety planning, data collection, evaluation and reporting during drug, biologic and vaccine development: a report of the Safety Planning, Evaluation and Reporting Team (SPERT). Clin Trials 2009; 6: 430-440. http://dx.doi.org/10.1177/1740774509344101
21. Fernandez Teruel C, Nalda MR, Gonzalez Alvarez I *et al*. Computer simulations of bioequivalence trials: selection of design and analyte in BCS drugs with first-pass hepatic metabolism: linear kinetics (I). Eur J Pharm Sci 2009; 36: 137–146. http://dx.doi.org/10.1016/j.ejps. 2008.10.014
22. Shah VP, Midha KK, Findlay JWA *et al*. Bio analytical method validation—a revisit with a decade of progress. Pharm Res 2000; 17: 1551–1557. http://dx.doi.org/10.1023/A:1007669411738
23. Mallet CR, Lu Z, Mazzeo JR. A study of ion suppression effects in electro spray ionization from mobile phase additives and solid-phase extracts. Rapid Commun Mass Spectrom 2004; 18: 49–58. http://dx.doi. org/10.1002/rcm.1276
24. Kranz H, Guthmann C, Wagner T, Lipp R, Reinhard J. Development of a single unit extended release formulation for ZK 811 752, a weekly basic drug. European Journal of Pharmaceutical Sciences 2005; 26: 47–53. http://dx.doi.org/10.1016/j.ejps.2005.04.018
25. Stephenson GA, Aburub A, Woods TA. Physical stability of salts of weak bases in the solid-state. Journal of Pharmaceutical Sciences 2011; 100: 1607–1617. http://dx.doi.org/10.1002/jps.22405
26. Zheng X, Yang R, Zhang Y, Wang Z, Tang X, Zheng L. Part II: bioavailability in beagle dogs of nimodipine solid dispersions prepared by hot-melt extrusion. Drug Development and Industrial Pharmacy 2007; 33: 783–789. http://dx.doi.org/10.1080/03639040601050205
27. Yamashita K, Nakate T, Okimoto K, Ohike A, Tokunaga Y, Ibuki R, Higaki K, Kimura T. Establishment of new preparation method for solid dispersion formulation of tacrolimus. International Journal of Pharmaceutics 2003; 267: 79–91. http://dx.doi.org/10.1016/j.ijpharm. 2003.07.010
28. Zheng X, Yang R, Zhang Y, Wang Z, Tang X, Zheng L. Part II: bioavailability in beagle dogs of nimodipine solid dispersions prepared by hot-melt extrusion. Drug Development and Industrial Pharmacy 2007; 33: 783–789. http://dx.doi.org/10.1080/03639040601050205
29. Bak A, Gore A, Yanez E, *et al*. The co-crystal approach to improve the exposure of a water-insoluble compound: AMG 517 sorbic acid co-crystal characterization and pharmacokinetics. Journal of Pharmaceutical Sciences 2008; 97: 3942–3956. http://dx.doi.org/ 10.1002/jps.21280
30. Horter D, Dressman JB. Influence of physicochemical properties on dissolution of drugs in the gastrointestinal tract. Advanced Drug Delivery Reviews 2001; 46: 75–87.
31. Gomez Orellana I. Strategies to improve oral drug bioavailability. Expert Opin Drug Deliv 2005; 2: 419-433. http://dx.doi.org/10.1517/ 17425247.2.3.419
32. Ettmayer P, Amidon GL, Clement B, Testa B. Lessons learned from marketed and investigational prodrugs. J Med Chem 2004; 47: 2393-2404. http://dx.doi.org/10.1021/jm0303812
33. Porter JH, Charman WN. *In vitro* assessment of oral lipid based formulations. Adv. Drug Deliv. Rev 2001; 50: 127–147. http://dx .doi.org/10.1016/S0169-409X(01)00182-X
34. Amidon GL, Lennernäs H, Shah VP, Crison JR. A theoretical basis for a biopharmaceutic drug classification: the correlation of *in vitro* drug product dissolution and *in vivo* bioavailability. Pharm. Res 1995; 12: 413– 420. http://dx.doi.org/10.1023/A:1016212804288

Source of support: Nil, Conflict of interest: None Declared

QUICK RESPONSE CODE	ISSN (Online) : 2277 –4572
	Website http://www.jpsionline.com

How to cite this article:
Balkrishana D. Tiwari, Omkar N. Shikare, Amruta M. sontakke. Bioequivalence study: Overview. J Pharm Sci Innov. 2014;3(5):421-424 http://dx.doi.org/10.7897/2277-4572.035187