Tropical Journal of Pharmaceutical Research September 2018; 17 (9): 1685-1691
ISSN: 1596-5996 (print); 1596-9827 (electronic)

Available online at http://www.tjpr.org
http://dx.doi.org/10.4314/tjpr.v17i9.1

Original Research Article

Comparative *in vitro* dissolution and *in vivo* bioavailability of commercial amlodipine tablets

Helgi Jung-Cook[1], Lourdes Mayet-Cruz[1], María Elena Girard-Cuesy[2]
[1]Departamento de Farmacia, Facultad de Química, Universidad Nacional Autónoma de México. Ciudad de México, [2]Investigación, Ciencia y Tecnología (ICT) Internacional, México

For correspondence: **Email: helgi@unam.mx; **Tel:** +52 55 56 22 52 82; **Fax:** +52 55 56 22 53 29*

Sent for review: 30 November 2017 *Revised accepted: 24 August 2018*

Abstract

Purpose: *To evaluate the in vivo and in vitro behavior of amlodipine immediate release products.*
Methods: *Three Mexican amlodipine products and the innovator (Norvasc®) were evaluated. Three bioequivalence studies were performed in 24 healthy male and female volunteers each. Plasma concentrations were determined using a liquid chromatographic method coupled with tandem mass spectrometry (LC/MS/MS). Dissolution profiles were evaluated using USP type apparatus 2 at 75 rpm and 500 mL of HCl 0.1N, pH 4.5 and pH 6.8. Also, the dissolution behavior of different lots of the innovator product was evaluated using apparatus 1 or 2 and 900 mL of buffer pH 6.8.*
Results: *All the generic products under study were bioequivalent to the innovator. In vitro data showed that although at pH 1.2 and 4.5, the products met the specifications for very rapidly dissolving products but at pH 6.8, neither the innovator nor the test products complied with the criteria for rapidly dissolving products. When the study was performed at pH 6.8 in 900 mL of medium, the innovator showed a rapid dissolution behavior.*
Conclusion: *The results show that the use of WHO conditions (900 mL of media, apparatus 2 at 75 rpm) are more adequate to predict the in vivo behavior of the amlodipine products.*

Keywords: *Biopharmaceutics Classification System (BCS), Dissolution, Bioequivalence, Solubility, Permeability*

Tropical Journal of Pharmaceutical Research is indexed by Science Citation Index (SciSearch), Scopus, International Pharmaceutical Abstract, Chemical Abstracts, Embase, Index Copernicus, EBSCO, African Index Medicus, JournalSeek, Journal Citation Reports/Science Edition, Directory of Open Access Journals (DOAJ), African Journal Online, Bioline International, Open-J-Gate and Pharmacy Abstracts

INTRODUCTION

The Biopharmaceutics Classification System (BCS) is used as a waiver of bioequivalence for immediate-release solid dosage forms [1]. Although the different BCS guidelines agree that the biowaiver criteria can be applied to BCS class 1 or class 3 drug products, there is no international consensus on the test conditions. Thus, the Guideline for the Investigation of Bioequivalence, EMA [2] specifies the following experimental conditions: apparatus 1 at 100 rpm or apparatus 2 at 50 rpm, using 900 mL or less and the following dissolution media: pH 1.0 – 1.2 (usually 0.1 N HCl or SGF without enzymes), pH 4.5 and pH 6.8 (or SIF without enzymes at 37 ± 0.5 ºC). The same conditions are specified in the Health Canada Biowaiver Guidance [3] however this guidance indicates that if coning is observed for the test as well as for the reference products,

speed could be increased to 75 rpm, nevertheless, the results obtained with the lower speed should also be reported. WHO guidance indicates that the test should be performed in 900 mL or less of dissolution media at pH 1.2, 4.5 and 6.8, using apparatus 2 at 75 rpm or apparatus 1 at 100 rpm [4], while FDA Guidance [5] recommends the use of apparatus 2 at 75 rpm or apparatus 1 at 100 rpm, and 500 mL of buffer media at pH 1.2, 4.5 and 6.8.

Amlodipine besylate is a long acting calcium channel blocker dihydropyridine used for treatment of hypertension, coronary artery disease and chest pain (angina) [6,7]. Amlodipine is indexed in the Essential Medicines WHO Model List as an antihypertensive drug in dose of 5 mg [8]. In Mexico, it is also included in the Basic Drug Catalog as 5 mg tablets [9]. The recommended dose is 5 - 10 mg once daily. Amlodipine is a weak base with a pKa value of about 9.0 at 25 ºC. The reported aqueous solubility is 0.774 mg/mL [10]. In relation to amlodipine besylate, solubility values at pH 1.2, 4.5 and 6.8 are 0.38 ± 0.017, 0.31 ± 0.005 and 0.110 ± 0.002 mg/mL respectively [11]. Since the dose administered is up to 10mg, the dose-to-solubility ratio is low, and therefore it is considered a highly soluble compound. With regard to its permeability, Caron *et al* [12] found that neutral amlodipine shows high permeability while cationic amlodipine does not permeate. *In vivo* studies have shown that although bioavailability is low (60 – 65 %), its permeability could be considered high due to metabolite excretion in urine (90 – 95 %) [13]. Moreover, the WHO guidance assigned amlodipine to BCS class 1 [4]; therefore, it has been considered as a candidate for a biowaiver through dissolution testing.

The main purposes of the present study were to assess the bioequivalence of three marketed products of amlodipine and to evaluate the effect of different media in the dissolution profile of these products.

EXPERIMENTAL

Materials and reagents

Amlodipine besylate and dexamethasone (internal standard) were acquired from Sigma-Aldrich. HPLC organic solvents were obtained from J.T. Baker. Drug release media and buffers were prepared using hydrochloric acid, acetic acid, sodium acetate, potassium chloride and potassium dihydrogenphosphate (J.T. Baker). Water was obtained from a Milli-Q (Millipore, Milford, MA, USA) system.

Drug products

Two different batches of the innovator product (Norvas® 5 mg) (Pfizer, Mexico) (R1 and R2) and three generic products containing 5 mg of amlodipine besylate (B, C and D) marketed in Mexico were evaluated.

In vivo studies

Three separate bioequivalence studies, in 24 healthy male and female volunteers each group, were performed using the innovator product (Norvas® 5 mg) as the reference product. The studies were conducted in accordance to the Helsinki Declaration [14]. Protocols [number BE0734, BE06026 and BE13021] were approved by the Ethics Committee (Comité de Ética e Investigación Biofarmacéutico de México). All the subjects gave their written informed consent prior to study admission.

Each Bioequivalence study was performed using a randomized, cross-over design 2 x 2 with 2-week washout.

In the first two bioequivalence studies (using products B and C), each subject received a single oral dose of 10 mg (two 5 mg tablets) of the reference (R1) or the test product. In the third study, a single 5-mg dose of the reference (R2) or of test product (D) was administered.

Products were orally administered after 10 h fasting with 250 mL of water. No food intake was permitted for 4 h after dosing. At this time, a standard meal was provided. Blood samples were taken at pre-dose and at the following times: 1, 2, 3, 4, 6, 8, 10, 12, 14, 24, 48, 96, 120 and 144 h. Samples were centrifuged at 3000 rpm for 10 min. Plasma was separated and kept at - 70 °C until assay.

Plasma concentrations of amlodipine were determined using a liquid chromatographic method with tandem mass spectrometry (LC-MS/MS), which was developed and validated before the studies were performed. The system consisted of a Shimadzu SIL-HTA autosampler (Kyoto, Japan) coupled to a turbo ionspray ionization-triple quadrupole mass spectrometer API 4000 (AB MDS Sciex. Toronto, Canada), with positive ion electrospray ionization using multiple reaction monitoring (MRM) mode. The analytical column was a Gemini® (Phenomenex) C_{18} (5 µm, 150 mm x 4.6 i.d.). The mobile phase consisted of acetonitrile:methanol (70:30, v/v) with 20 mM of ammonium acetate. Table 1 shows tandem mass spectrometric parameters.

Table 1: Tandem mass spectrometric parameters of amlodipine and dexamethasone

Compound	Mol Wt (g/mol)	Protonated ion	Fragment	*CE (eV)*	*DP (V)*	*EP (V)*	*CXP (V)*
Amlodipine	408.879	409.224	237.939	13.49	41.48	10	4.99
Dexamethasone (internal standard)	392.464	393.214	373.200	14	35.54	6.8	6.7

Note: *CE* = Collision energy, *eV* = Electron volt, *DP* = Declustering potential, *V* = Volt, *EP* = Entrance potential, *CXP* = Collision cell exit potential

Sample preparation: A volume of 400 µL of plasma sample was transferred to an assay tube and spiked with 100 µL of internal standard (dexamethasone) at a concentration of 250 ng/mL. Then 200 µL of 1M sodium hydroxide were added mixed in vortex during 1 min and 3 mL of ether:hexane:dichloromethane (60:30:10) were added. Samples were shaken in vortex and centrifuged. The organic layer was evaporated with nitrogen stream at 40 °C. The residue was reconstituted using 300 µL of mobile phase and 20 µL were injected into the chromatographic system. The analytical assay was linear from 0.1 – 12 ng/mL. Intra-day and inter-day coefficients of variation were less than 15 %. The recovery from amlodipine ranged from 80 to 85%.

Non compartmental analysis of pharmacokinetic parameters was performed using WinNonlin Version 5.0.1 (Pharsight, Mountain View, CA, USA). The following pharmacokinetic parameters were obtained: C_{max}, t_{max}, area under the curve from 0 to the time of the last measurable plasma concentration ($AUC_{0 - tlast}$), AUC extrapolated to time infinity ($AUC_{0 - inf}$) and terminal elimination half-life (t1/2).

Bioequivalence was established if the 90 % confidence intervals of the geometric mean ratios of the plasma C_{max} and AUC fell within the range of 80 % to 125 %.

In vitro studies

Dissolution profiles were evaluated using the same products as in the bioequivalence studies. Studies were carried out using USP apparatus 2 (Vankel 7000) at 75 rpm with twelve replicates at 37 ± 0.5 °C. The following media (500 mL) were used: pH 1.2 (hydrochloric acid solution), 0.05 M acetate buffer pH 4.5, and 0.05 M phosphate buffer pH 6.8. In all cases, 5 mL samples were removed at 10, 15, 20, 30 and 45 min without medium reposition. Samples were filtered through 0.45 µm HV Durapore® membrane filters (Millipore) and assayed using a previously validated spectrophotometric method at 240 nm (UV/VIS Shimadzu spectrophotometer). The method was linear from 1 - 12 µg/mL. Intra-day and inter-day coefficients of variation were < 2 %. For dissolution profile comparison, f_2 similarity test was used.

Additionally, the dissolution profiles of other three batches of the reference product: R3, R4 and R5 were evaluated using 900 mL of pH 6.8 buffer and apparatus 2 at 75 rpm or apparatus 1 at 100 rpm.

Solubility study at pH 6.8

Amlodipine besylate, equivalent to 5 mg of amlodipine, was placed in 250 mL of phosphate buffer pH 6.8. Samples were stirred at 37 °C for 48 h. Afterwards, samples were filtered using a 0.45 µm HV Durapore® membrane filters (Millipore) and analyzed with the same analytical method used for the dissolution study. The experiment was performed in triplicate.

RESULTS

In vivo data

Bioequivalence studies were conducted in accordance with the current bioequivalence guidelines [15,16]. In each bioequivalence trial, 24 subjects were enrolled and completed the study and no severe adverse events occurred during the different studies.

Figure 1 shows the mean plasma concentration data of amlodipine obtained in the three bioequivalence studies. It can be seen that amlodipine plasma profiles were similar between the test and the reference products, with peak levels around 6 h after drug administration.

Table 2 shows the mean values of the pharmacokinetic parameters, as well as the 90 % confidence intervals for the different amlodipine products under study. Results showed that the mean half-life was approximately 49 hours for all treatments. In the three bioequivalence studies, C_{max}, $AUC_{0 - 144\ h}$ and $AUC_{0 - inf}$ for each pair of products (test *vs* reference) were not statistically different ($p > 0.05$). The relative bioavailability (F) was 0.99, 1.03 and 1.03 for the ratios B/R1, C/R1 and D/R2, respectively. Moreover, the 90 % confidence intervals of the log transformed $C_{max,}$

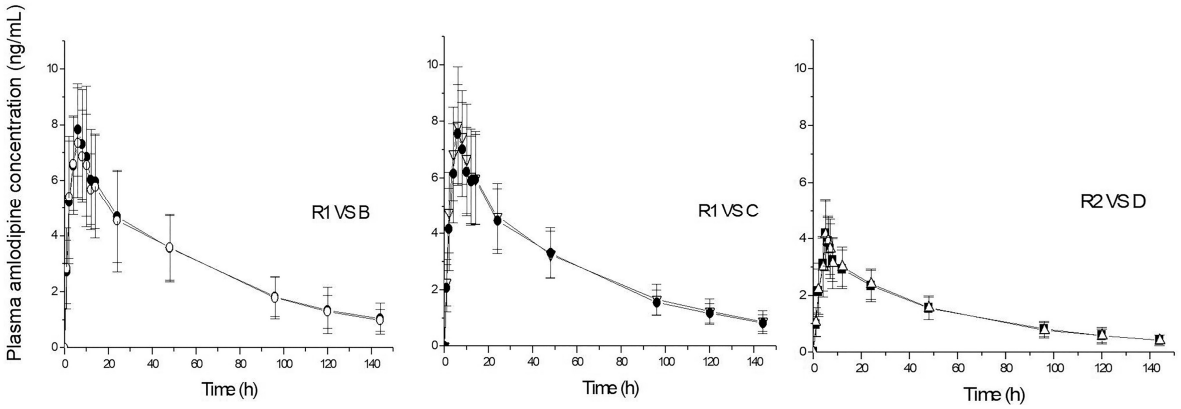

Figure 1: Plasma profiles of amlodipine besylate after the administration of the different products in bioequivalence studies, (R1 VS B and R1 VS C single oral dose of 10 mg, R2 VS D single oral dose of 5 mg). ***Keys:*** ● R1, ○ B, ▽ C, △ D, ■ R2

$AUC_{0-144\ h}$ and AUC_{0-inf} in each set of studies were within the range of 80 – 125 %. On the basis of the above analysis, the test products were considered bioequivalent with the innovator product. In the different studies, the intra-subject coefficient of variation for C_{max} was 11.4, 10.9 and 11.6 %, while the values for AUC_{0-144h} were 15.4, 12.7 and 10.9 % (R1 *vs* B, R1 *vs* C and R2 *vs* D), respectively.

In vitro results

Figure 2 illustrates the release properties of the amlodipine besylate products in the different dissolution media. It can be seen that at pH 1.2 and 4.5, all the products fulfilled the criteria for very rapidly dissolving products (> 85 % dissolved within 15 min). Nevertheless when pH 6.8 was used, neither the reference nor the test products B and C met the criteria for rapidly dissolving products (> 85 % dissolved within 30 min). Under these dissolution conditions, all the test products had a higher dissolution rate than the reference product and therefore none of them met the (f_2) acceptance criteria with values of 37, 41 and 39 for products B, C and D, respectively.

Due to the low percentage of amlodipine dissolved at pH 6.8 for most of the products, a solubility study of the drug substance was performed. The results showed that the dose of 5 mg of amlodipine is completely soluble at this pH.

Table 2: Mean pharmacokinetic parameters and confidence Intervals using ln-transformed data in the three bioequivalence studies after oral administration of amlodipine besylate

R1 *vs* B	**R1 Mean±SD)**	**B Mean±SD**	**Confidence Intervals**	**Power**
Single oral dose (10mg)				
C_{max} ($ngmL^{-1}$)	8.4±2.3	8.3±2.0	93.24-104.37	0.99
$AUC_{0\rightarrow tlast}$($nghmL^{-1}$)	419.6±44.7	415.8±128.6	93.68-109.11	0.99
$AUC_{0\rightarrow inf}$($nghmL^{-1}$)	499.1± 93.4	479.8±158.5	92.22-102.98	0.99
t_{max} (h)	7.3±2.4	7.1±1.6		
$t_{1/2}$ (h)	48.4±14.1	45.5±12.2		
R1 *vs* C	**R1**	**C**		
Single oral dose (10mg)				
C_{max} ($ngmL^{-1}$)	7.8±1.8	8.3±1.8	96.83-107.91	0.99
$AUC_{0\rightarrow tlast}$ ($nghmL^{-1}$)	398.8±86.8	409.2±101.1	99.30-112.63	0.99
$AUC_{0\rightarrow inf}$($nghmL^{-1}$)	459.7±103.2	476.3±131.4	97.53-106.17	0.99
t_{max} (h)	7.4±2.8	7.4±3.1		
$t_{1/2}$ (h)	49.4±16.9	47.6±11.9		
R2 *vs* D	**R2**	**D**		
Single oral dose (5mg)				
C_{max} ($ngmL^{-1}$)	4.5 ±1.1	4.4±1.2	93.04-104.36	0.99
$AUC_{0\rightarrow tlast}$ ($nghmL^{-1}$)	198.6± 46.1	200.5±46.5	96.03-105.70	0.99
$AUC_{0\rightarrow inf}$($nghmL^{-1}$)	234.6±66.5	232.2 ± 61.6	93.85-104.50	0.99
t_{max} (h)	5.6±0.8	5.5 ± 0.8		
$t_{1/2}$ (h)	51.0±16.7	48.7 ± 11.5		

Table 3: Drug dissolved at pH 6.8 (mean ± RSD) in batches of reference product (R3, R4 and R5) at different conditions of agitation, media volume and apparatus

Variable	% Dissolved at pH 6.8	R3 (mean ± RSD)	R4 (mean ± RSD)	R5 (mean ± RSD)
Apparatus 2, 500 mL, 75 rpm	15 min	68±4	63±10	58±4
	30 min	79±3	73±8	68±4
Apparatus 2, 900 mL, 75 rpm	15 min	79±7	76±3	78±3
	30 min	87±6	86 ±3	86±3
Apparatus 1, 900 mL, 100 rpm	15 min	91±2	94±4	88±1
	30 min	95±3	99±4	93±3

RSD: Relative Standard Deviation

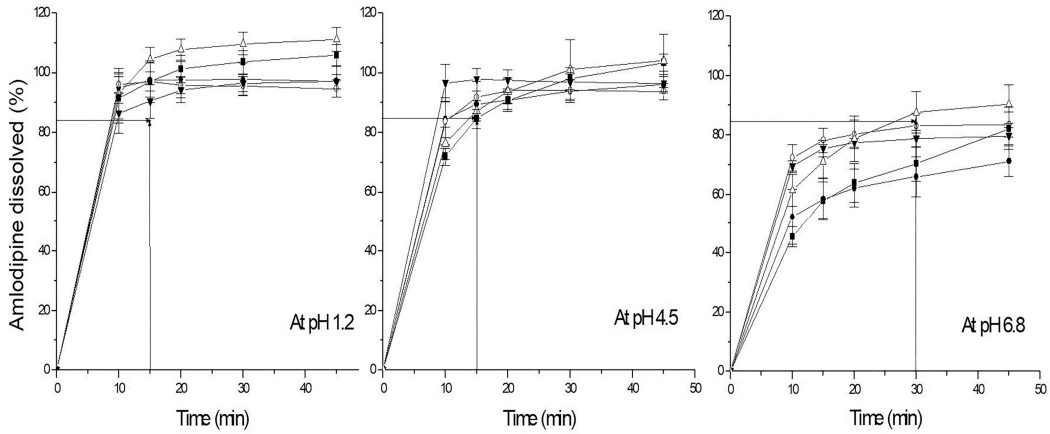

Figure 2: Release profiles of amlodipine in HCl 0.1N, pH 4.5 and pH 6.8.
Keys: ● R1, ○ B, ▽ C, △ D, ■ R2

Taking into account the low dissolution of the reference product at pH 6.8, we decided to compare the performance of three additional batches (R3, R4 and R5) in this medium, using apparatus 2 at 75 rpm and 500 or 900 mL of dissolution media or apparatus 1 at 100 rpm and 900 mL of media. Table 3 shows the results obtained. It can be see that when 500 mL of media were used, the dissolution behavior was consistent with those previously obtained with batches R1 and R2, with low percentage dissolved at 30 min. Nevertheless when 900 mL of media and apparatus 2 was used, batches complied with the rapidly dissolution criteria. In the case of apparatus 1, the products dissolved more than 85 % in 10 minutes, and therefore complied with the acceptance criteria for very rapidly dissolving products.

DISCUSSION

The Biopharmaceutical Classification System (BCS) provides a scientific framework to determine either the requirement of bioequivalence studies for generic products or if *in vitro* data can be applied to support a waiver for an *in vivo* study [17,18]. With regard to amlodipine, different studies have been performed to determine the release of amlodipine Besylate tablets using various dissolution conditions, and results are discordant. Thus, Shohin *et al* [13] evaluated the dissolution characteristics of the reference product and one product marketed in Russia, using 500 mL of USP buffer solutions at pH 1.2, 4.5 and 6.8 and apparatus 2 at 75 rpm. They found that the products dissolved very rapidly at pH 1.2 and 4.5, while at pH 6.8 they behaved as rapidly dissolving products and dissolution profiles were comparable.

Akinleye *et al* [19] evaluated the dissolution profiles of two generic products of amlodipine available in Nigeria, and the reference product (Norvasc®) using apparatus 2 at 50 rpm and 900 mL of buffers at pH 1.2, 4.5 and 6.8. They found that in all media, dissolution was low, and hence none of the products complied the biowaiver criteria for very rapidly or rapidly dissolving tablets. The f_2 test showed that the release of one of the generic products (B) was similar to the innovator product in all media ($f_2 \geq 50$), while the similarity factor f_2 of the other generic product (A) at pH 4.5 was lower than 50 due to the rapid dissolution of the generic product. On the other hand, Feroz *et al* [20] evaluated the dissolution

characteristics of six different brands of amlodipine besylate 5 mg tablets in 900 mL of different dissolution media (water, pH 1.2, 4.5 and 6.8) using apparatus 2 at 75 rpm. They found that under biowaiver conditions, all products tested behaved as very rapidly dissolving products, and therefore they were considered equivalent.

To our knowledge, this is the first report in which the *in vivo* as well as the *in vitro* characteristics of amlodipine besylate under biowaiver conditions were evaluated. *In vivo* studies showed that after the oral administration of amlodipine, C_{max} and t_{max} values were similar to those reported [21,22]. The results also revealed that the three generic products were bioequivalent to the innovator product; however, the *in vitro* dissolution profiles at pH 6.8 using 500 ml of media were not able to predict the *in vivo* performance of the products, since the products neither complied with the biowaiver criteria for rapidly dissolving products nor the criteria for similar dissolution profiles. The results obtained with several batches of the innovator product under different dissolution conditions showed that although amlodipine is a class 1 drug, the release is influenced by the volume of medium as well as by the dissolution apparatus. FDA Draft Guidance [9] recommends that, for biowaivers, dissolution studies should be carried out using apparatus 2 at 75 rpm in 500 mL of medium at pH 1.2, 4.5 and 6.8. Our results show that with this media volume, dissolution test at pH 6.8 is too sensitive, resulting in a false characterization of the products. Data also show that using apparatus 2 at 75 rpm in a volume of 900 mL could be an indicator of the bioavailability of amlodipine.

CONCLUSION

Amlodipine is classified as a BCS class 1 drug and could be elegible for a biowaiver based approval. The generic products evaluated are bioequivalent with the innovator. The *in vitro* results indicate that although at pH 1.2 and 4.5, all the products complied with the very rapid dissolution criteria, but differences were found when the dissolution was performed at pH 6.8. WHO biowaiver conditions (900 mL of media at ph 6.8, apparatus 2 at 75 rpm) were more adequate to predict the rate and extent of the dissolution of the amlodipine products.

DECLARATIONS

Acknowledgement

This research was partially supported by Programa de Apoyo a la Investigación y al Posgrado (PAIP 50009137), Facultad de Química, UNAM.

Conflict of interest

No conflict of interest is associated with this work.

Contribution of authors

We declare that this work was done by the author(s) named in this article and all liabilities pertaining to claims relating to the content of this article will be borne by the authors. All authors read and approved the manuscript for publication.

REFERENCES

1. *Amidon GL, Lennernäs H, Shah VP, Crison JR. A Theoretical Basis for a Biopharmaceutics Drug Classification: The correlation of in vitro drug product dissolution and in vivo bioavailability. Pharm Res 1995; 12: 413-420.*
2. *Committee for Medicinal Products for Human Use (CHMP), European Medicines Agency (EMA) 2010. Guideline on the Investigation of Bioequivalence.*
3. *Bureau of Policy, Science and International Programs. Therapeutic Products Directorate, Health Canada 2014. Biopharmaceutics Classification System Based Biowaiver.*
4. *WHO Technical Report Series, No. 937. World Health Organization; 2006, Annex 8. Proposal to waive in vivo bioequivalence requirements for WHO Model List of Essential Medicines immediate-release, solid oral dosage forms.*
5. *The US Food and Drug Administration (FDA), 2015. FDA Center for Drug Evaluation and Research. U.S. Department of Health and Human Services. Guidance for Industry: Waiver of in vivo Bioavailability and Bioequivalence Studies for Immediate-Release Solid Oral Dosage Forms Based on a Biopharmaceutical Classification System. DRAFT GUIDANCE. Biopharmaceutics Revision 1 [cited 2017 Jan 10]. Available from: https://www.fda.gov/downloads/Drugs/Guidances/ucm070246.pdf*
6. *Pfizer, 2006. Amlodipine besylate prescribing information.*
7. *Drugs.com ©2000-2017 [cited 2017 Apr 28]. Available from: http://www.drugs.com/monograph/amlodipine-besylate.html*
8. *WHO Model List of Essential Medicines, 2017. World Health Organization; 20th List. [Updated: 2017 March; cited 2017 Apr 30]. Available from: http://www.who.int/medicines/publications/essentialmedicines/20th_EML2017.pdf?ua=1*
9. *Consejo de Salubridad General, MEXICO, 2016. Cuadro Básico y Catálogo de Medicamentos. [cited 2017 Jan*

30] Available from: http://www.csg.gob.mx/descargas/pdfs/cuadro_basico/CB2014/index/EDICION_2016_MEDICAMENTOS.pdf

10. Austin RP, Davis AM, Manners CN. Partitioning of ionizing molecules between aqueous buffers and phospholipid vesicle. J Pharm Sci 1995; 84(10): 1180-1183.
11. Pant T, Mishra K, Subedi RK. In vitro studies of amlodipine besylate tablet and comparison with foreign brand leader in Nepal. Int J Pharm Sci Drug Res 2013; (10): 3958-3964.
12. Caron G, Ermondi G, Damiano A, Novaroli L, Tsinman O, Ruell JA, Avdeef A. Ionization, lipophilicity and molecular modeling to investigate permeability and other biological properties of amlodipine. Bioorg Med Chem 2004; 12: 6107-6118.
13. Shohin IE, Ramenskaya GV, Vasilenko GF, Malashenko EA. In vitro dissolution kinetics of amlodipine tablets marketed in Russia under biowaiver conditions. Dissolut Technol 2010;17: 20-22.
14. Declaration of Helsinki. Bulletin of the World Health Organization, 2001;79(4) [homepage on the internet] [cited 2018 Apr 04] Available from: http://www.who.int/bulletin/archives/79%284%29373.pdf
15. Estados Unidos Mexicanos-Secretaría de Salud, 2013. NOM-177-SSA1-2013, Que establece las pruebas y procedimientos para demostrar que un medicamento es intercambiable. Requisitos a que deben sujetarse los Terceros Autorizados que realicen las pruebas de intercambiabilidad. Requisitos para realizar los estudios de biocomparabilidad. Requisitos a que deben sujetarse los Terceros Autorizados, Centros de Investigación o Instituciones Hospitalarias que realicen las pruebas de biocomparabilidad.
16. The US Food and Drug Administration (FDA), 2013. FDA Center for Drug Evaluation and Research. U.S. Department of Health and Human Services. Guidance for Industry: Bioequivalence Studies with Pharmacokinetic Endpoints for Drugs Submitted Under an ANDA. DRAFT GUIDANCE. Biopharmaceutics [cited 2017 Jan 10]. Available from: https://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM377465.pdf
17. Anand O, Yu LX, Conner DP, Davit BM. Dissolution Testing for Generic Drugs: An FDA Perspective. AAPS J 2011 Sep; 13(3): 328–35.
18. Committee for Medicinal Products for Human Use (CHMP), European Medicines Agency (EMEA), 2007. Concept paper on BCS-Biowaiver, EMEA/CHMP/EWP/213035/2007.
19. Akinleye MO, Oyetunde OO, Okpara HE, Ayerota EL. Equivalence of two generic brands of amlodipine besylate under biowaiver conditions. Int J Pharm Pharm Sci 2012; 4: 265-68.
20. Feroz M, Razvi N, Ghayas S, Anjum F, Ghazal L, Siddiqui SA. Assessment of pharmaceutical quality control and equivalence of various brands of amlodipine besylate (5 mg) tablets available in the Pakistani market under biowaiver conditions. Int J Pharm Pharm Sci 2015; (2): 909-913.
21. Carvalho M, Oliveira CH, Mendes GD, Sucupira M, Moraes ME, De Nucci G. Amlodipine Bioequivalence Study: Quantification by liquid chromatography coupled to Tandem Mass Spectrometry. Biopharm Drug Dispos 2001; 22: 383–290.
22. Park JY, Kim KA, Lee GS, Park PW, Kim SL, Lee YS, Lee YW, Shin EK. Randomized, open-label, two-period crossover comparison of the pharmacokinetic and pharmacodynamic properties of two amlodipine formulations in healthy adult male Korean subjects. Clin Ther 2004 May; 26(5): 715-723.