dx.doi.org/10.14227/DT280121PGC1

Comparative Assessment of Critical Quality Attributes of Sildenafil Tablets

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ABSTRACT

Sildenafil citrate is a selective inhibitor of the enzyme phosphodiesterase type 5, used to treat erectile dysfunction in adults and pulmonary hypertension, mainly in children. This work aimed to perform a comparative study of sildenafil tablets marketed in Argentina and establish their pharmaceutical equivalence. Eight commercial formulations (immediate-release tablets) containing 50 mg of sildenafil were analyzed according to United States and Argentinian Pharmacopoeial guidelines. The assay was performed by UV spectrophotometry in 0.01 N hydrochloric acid. Similar conditions were used for dissolution tests, which were carried out in a basket apparatus at 100 rpm. All samples met pharmacopeial specifications for acceptance (i.e., assay, content uniformity, hardness, friability, disintegration, and in vitro dissolution) for immediate-release dosage forms. When compared to the reference formulation, a statistically significant difference was noted for dissolution efficiency in one case (sample F). Based on the obtained results, it is possible to conclude that the evaluated formulations of sildenafil can be considered pharmaceutical equivalents.

KEYWORDS: Dissolution profiles, pharmaceutical equivalence, sildenafil tablets

INTRODUCTION

Sildenafil citrate is a selective inhibitor of the phosphodiesterase (PDE) type 5 enzyme, widely used for erectile dysfunction treatment in adults (*1, 2*). The drug was first approved by the U.S. Food and Drug Administration (FDA) in March 1998 after 2 years of being patented; the drug was initially proposed to treat hypertension and angina pectoris (*3*). Later in 2005, the FDA approved the application of sildenafil citrate to treat pulmonary hypertension in adults and pediatric patients (*2, 4*). Different provisional biopharmaceutical classifications for sildenafil citrate have been described in the literature, and Miranda et al. confirmed that sildenafil citrate can be classified as a class II drug in the Biopharmaceutical Classification System (BCS) based on solubility and permeability data (*1*). According to the criteria of World Health Organization (WHO), FDA, and European Medicines Agency (EMA), the BCS-based biowaiver procedure cannot be recommended for immediate-release formulations of sildenafil citrate, and its interchangeability would not be fully guaranteed through in vitro similarity studies (*1*).

The WHO indicates that products can be considered pharmaceutical equivalents if they contain the same amount of active substance, use the same route of administration, and meet identical or comparable quality standards (*5*). The Argentinian pharmaceutical market is not depicted by generic drugs in a strict sense; rather, sildenafil immediate-release solid oral dosage forms are offered as one reference product and several multisource products. Patients frequently replace

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the reference product with a multisource formulation for economic reasons, regardless of quality.

In 1998, sildenafil citrate was only commercialized by the pharmaceutical laboratory that patented it, Pfizer. Today, it is offered by more than 30 laboratories in Argentina. Despite it being a prescription medicine, evidence indicates that sildenafil is sold without a prescription. According to the Argentine Pharmaceutical Confederation (COFA), in 2013 almost 3.6 million sildenafil tablets were sold in pharmacies in Argentina, and another 3 million units were sold on the Internet and illegal sites (e.g., convenience stores, hotels, motels) (*6*).

Since consumers of sildenafil tablets in Argentina interchange formulations without any control, it became crucial to compare commercial products, at least in technical terms. To the extent of our knowledge, very few studies have been published in the literature concerning pharmaceutical equivalence of sildenafil citrate products, and none regarding samples from the Argentine market. A study in Costa Rica compared the innovator with a generic product in terms of dosage form uniformity (*7*). Similarly, a study performed in Nigeria compared nine products, but only in terms of sildenafil citrate content (*8*). Another study performed by Deconinck et al. in Belgium evaluated the reference product and 17 counterfeit products containing sildenafil citrate, including tablets, capsules, and an oral gel (*9*). Thus, the purpose of the present work was to assess and compare the critical quality attributes, including in vitro dissolution, for the pharmaceutical characterization of sildenafil tablets from the Argentine market and evaluate their pharmaceutical equivalence.

MATERIALS AND METHODS

Chemicals and Reagents

Sildenafil citrate was purchased from Saporiti (Parafarm, Argentina). Analytical grade hydrochloric acid (HCl) (Anedra, Argentina) was used to evaluate sildenafil concentration in different tests (i.e., assay, content uniformity, and in vitro dissolution).

Sildenafil Products

Eight commercial formulations, i.e., immediate-release coated tablets containing 50 mg of sildenafil, randomly labeled from A to H, were included in this study. All multisource products corresponded to national industries, whereas the reference formulation (E) was imported from Mexico. The samples were obtained from local pharmacies in Bahia Blanca city (Buenos Aires province) and were analyzed within their shelf life.

Equipment

Sildenafil concentration was assessed during the assay and in vitro dissolution testing using a UV spectrophotometer (Varian Cary 50 Conc, Varian Instruments, Australia). Three testers, i.e., DGM02, FGMO2, and EGMO2 (Scout Electronics, Argentina), were used to measure hardness, friability, and disintegration, respectively. A dissolution tester (DT60, Erweka GmbH, Germany) was used for in vitro dissolution assessment. Materials and tablets were weighed using an electronic analytical balance (Acculab ALC-210.4M, USA).

Methods for Evaluation of Critical Quality Attributes

A comparative study of the information included in patient leaflets and labels of primary and

secondary packaging was included in the analysis to evaluate the extent of agreement with national legislation (*10, 11*).

Ten tablets from each product were randomly selected for evaluation of weight variation. The results were expressed as mean weight ± standard deviation (SD).

Argentine Pharmacopeia guidelines were followed for friability, hardness, and disintegration tests (*10*). First, 10 tablets from each product were weighed before and after the friability procedure (100 revolutions of the friability tester); results (weight loss) were expressed as percentage. The maximum acceptable result is a value not higher than 1% (*10*). Second, another 10 tablets from each product were evaluated using the hardness tester; results were expressed as the degree of force (in kp) required to break the tablet. Disintegration tests were performed in distilled water at 37.0 ± 2.0 °C using six tablets of each product. The results were expressed as the maximum time needed for complete disintegration of each tablet, which should be lower than 30 minutes (*10*).

For sildenafil content evaluation, an accurately weighed amount of powder, obtained from 10 weighed and ground tablets of each product, was dissolved using 0.01 N HCl. The filtered solution (0.45-µm pore-size nylon membrane filter, Gamafil, Argentina) was diluted with the same reagent, and the sildenafil concentration was assessed by spectrophotometric analysis at 292 nm (*12*). The UV methodology was previously compared with the official method, with no significant differences detected between results ($p > 0.05$) (*13*). Therefore, a sildenafil standard calibration curve ($y = 0.0299x - 0.0056$; $R^2 = 1$) was developed for content determination and used for the assay and uniformity tests (i.e., 10 tablets of each product were individually assayed).

Dissolution tests were performed according to United States Pharmacopeia (USP) (*13*). The applied conditions involved testing in an apparatus 1 (basket) at an agitation speed of 100 rpm, with an exact volume (900 mL) of dissolution medium (0.01 N HCl) at 37.0 ± 0.5 ºC. At pre-selected time intervals (5, 10, 15, and 20 min), aliquots were withdrawn, suitably filtered (0.45-µm pore-size nylon membrane filter, Gamafil), diluted, and assayed by UV spectrophotometry (292 nm). A standard calibration curve was concomitantly constructed ($y = 0.0298x - 0.0003$; $R^2 = 0.9999$; concentration range = 4.0–34.0 µg/mL) for the determination of dissolved sildenafil. The USP specification for dissolution tests states that not less than 80% (Q) of the labeled amount of sildenafil should dissolve within 15 min (*13*). In addition, a statistical comparison of dissolution efficiency (DE) was performed via analysis of variance (ANOVA). DE is defined as the ratio, in percentage, of the area under the curve obtained from the dissolution profile, with the total area of the rectangle considered as 100% dissolution for the same time interval (*14*).

RESULTS AND DISCUSSION

The comprehensive concept of pharmaceutical equivalence includes not only the properties of the active pharmaceutical ingredient and pharmaceutical form but also the instructions for use and storage (especially when these instructions are crucial for stability and shelf-life of the product). For this reason, the information presented on labels (primary and secondary packaging) and patient leaflets of the evaluated products was compared according to WHO guidelines and national regulations (*5, 10, 11*). The official monograph states for sildenafil tablets says to "Preserve in well-closed containers. Store at controlled room temperature" (*13*). The conditions

for packaging and storage declared by manufacturers of the evaluated formulations are presented in Table 1, where several differences were recorded. Only the labels of formulations A and D specified to "store in its original packaging." The label information matched the recommendations described in the leaflet. Products A, D, F, G, and H indicated a temperature range for storage, but products B, C, and E only referred to a maximum temperature. Moreover, it is essential that information declared in labels and leaflets is uniform between the different commercial products and ascertained by regulatory agencies to avoid misunderstandings among patients, care givers, and medical professionals.

Table 1. Information of Evaluated Products and Results of Physical Quality Control Tests of Sildenafil (50-mg) Tablets

Product	Price[a]	Storage Conditions[b]	Weight (mg)[c]	Hardness (kp)[c]	Disintegration time (s)[d]
A	41.95	Store at a room temperature between 15 and 30 °C, protected from light in original container	410.7 ± 3.7	14.9 ± 0.4	85
B	57.50	Store at room temperature not higher than 30 °C	365.1 ± 4.8	9.4 ± 0.8	26
C	50.00	Do not expose to temperatures above 30 °C	308.4 ± 1.4	13.0 ± 4.1	29
D	44.00	Store in original container between 15 and 30 °C and dry place and protected from light	315.1 ± 1.7	9.4 ± 0.2	97
E (Ref)	73.70	Store at a temperature below 30 °C	311.1 ± 4.2	14.0 ± 3.6	159
F	52.50	Store at room temperature, preferably between 15 and 30 °C	302.4 ± 4.1	19.2 ± 0.9	436
G	77.29	Store between 15 and 30 °C	418.7 ± 4.2	13.3 ± 0.7	114
H	56.00	Store at room temperature, between 15 and 30 °C	305.5 ± 4.4	11.0 ± 0.2	47

[a]Price per tablet in Argentine pesos at the time of analysis.
[b]Information presented in labels and leaflets.
[C]Mean ± SD
[d]Maximum time needed for complete disintegration of evaluated tablets.
Ref: reference formulation.

As mentioned earlier, in Argentina, economic reasons represent the major factor concerning the extensive use of multisource products and the interchangeability decisions taken by patients. Sildenafil products revealed a wide range of selling prices in this particular market; notably, products E (reference) and G were almost 80% higher than products A and D (Table 1).

The results for weight variation, friability, and disintegration tests are also shown in Table 1. Differences were recorded between the mean weights of each product, with results in ranging from 302.4 to 418.7 mg. These observations could be attributed to differences in qualitative and quantitative excipient composition according to each formula and manufacturing process, which

consequently affects the size and shape of each product, with no strict relation to differences in drug content or dissolution performance. Mean hardness results ranged from 9.4 to 19.2 kp, which indicated suitable mechanical behavior because the results were all above the acceptable value, 2.0 kp. All formulations complied with the friability test because a very slight loss of powder was recorded (< 0.5% in all cases, data not shown), with values below the maximum allowed (*10*). With respect to disintegration results, all formulations fulfilled the Argentine Pharmacopeia specifications, with values between 26 seconds and 17 minutes (*10*).

The assay results are shown in Table 2. Mean ± SD values ranged from 93.5% ± 4.3 (B) to 100.1% ± 2.6 (E, reference). The reference formulation and products A and F met USP specifications for dissolution in Stage 1, and the remaining products met the specifications in Stage 2 (Table 2). All of the evaluated formulations can be classified as "very fast dissolving" (i.e., ≥ 85% within 15 min). Therefore, all formulations fulfilled the official specifications for these critical quality attributes (i.e., assay, uniformity of dosage units, hardness, friability, disintegration, and in vitro dissolution) (*10, 13*).

Table 2. Assay, Uniformity, and Dissolution Results for Sildenafil (50 mg) Tablets

Product	Assay[a]	Uniformity of Dosage Units (Range / RSD)[b]	Dissolution Test at S1 Stage (Range / RSD)[c]	Dissolution Efficiency
A	98.1 ± 7.2	93.3–113.2 / 7.6	87–90 / 1.3	78.6 ± 1.0
B	93.5± 4.3	88.3–98.7 / 5.6	84–90 / 2.6	76.7 ± 1.3
C	94.5 ± 6.4	85.0–99.4 / 6.0	84–100 / 6.0	80.7 ± 5.3
D	98.3 ± 0.6	98.4–99.1 / 0.4	84–87 / 1.4	76.8 ± 1.2
E (Ref)	100.1 ± 2.6	98.2–101.0 / 1.5	86–93 / 2.9	78.7 ± 2.0
F	97.8 ± 0.7	98.0–98.8 / 0.4	85–91 / 2.4	73.1 ± 3.8
G	97.9 ± 1.4	97.5–98.4 / 0.4	82–92 / 3.5	78.2 ± 3.8
H	94.5 ± 2.0	95.2–96.2 / 0.5	79–92 / 6.6	75.7 ± 4.6

Data are mean percent of labeled amount ± SD unless otherwise noted.
[a]Specification for acceptance: 90.0–110.0%.
[b]Specification for acceptance: 85.0–115.0%; RSD < 6%.
[c]USP specification for acceptance: 80% (Q) in 15 min.
Ref: reference formulation; RSD: relative standard deviation; Q: amount of dissolved active pharmaceutical ingredient, specified in the individual monograph, expressed as a percentage of labeled content of the dosage unit.

The evaluation of in vitro dissolution is essential for the assessment of the formulation performance in terms of batch quality and similarity between commercial (multisource or reference) products. Dissolution profiles of sildenafil formulations were constructed based on the mean percentage (± SD) of labelled amount dissolved at each sampling time (Fig. 1). It can be seen in Figure 1 that samples B, C, D, and E reached maximum dissolution at 5 minutes, whereas samples A, G, and H reached the plateau at 10 minutes. At 5 minutes, all formulations showed

dissolved percentages above 85% except for product F, which dissolved 73% at that time, then reached its maximum at 15 min. This result was in accordance with the disintegration time of formulation F, which was the highest value obtained (Table 1). Furthermore, this product exhibited a higher hardness result compared to the other products (Table 1). Higher disintegration and hardness test results could be related to the lowest dissolution rate obtained for product F.

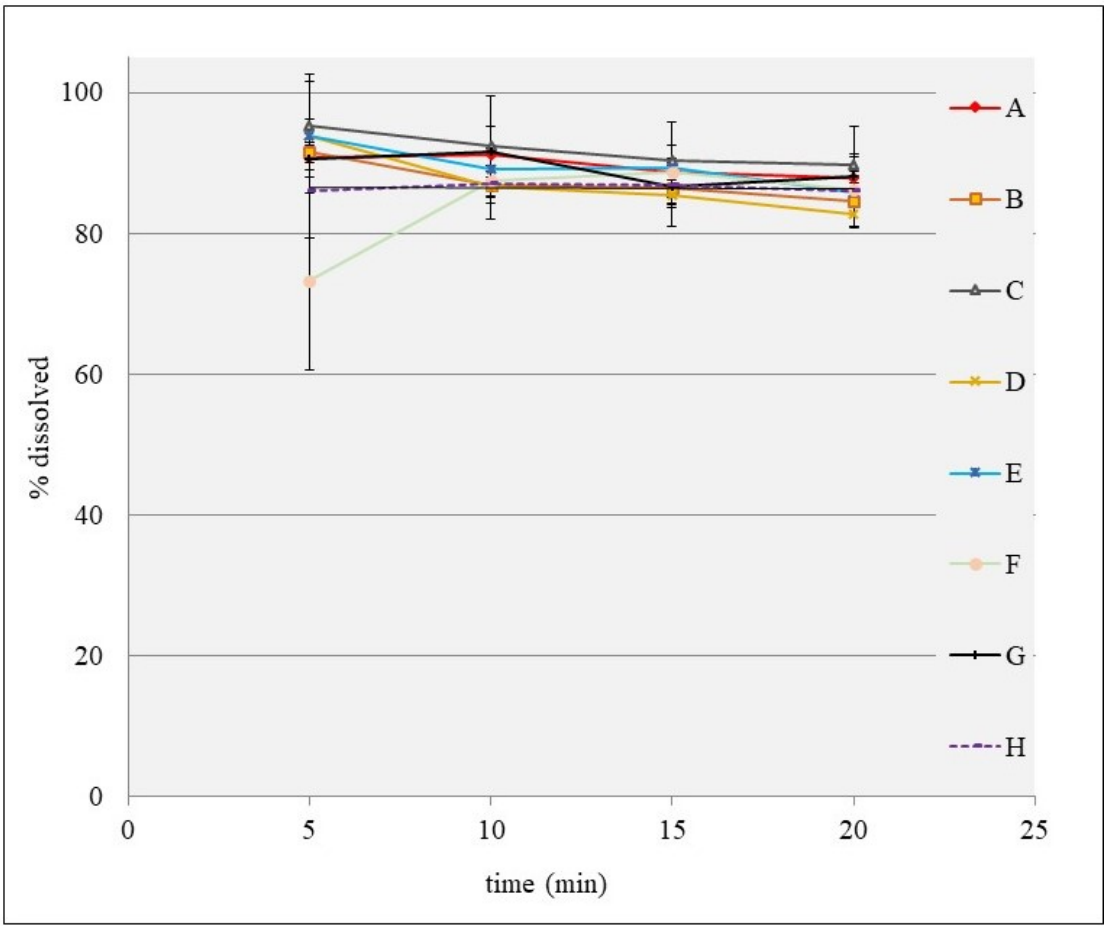

Figure 1. Dissolution profiles of sildenafil (50 mg) immediate-release tablet formulations (mean percentage of labeled amount dissolved ± SD); sample E is reference formulation.

Dissolution profiles were compared base on the DE parameter. If a formulation presents high values of DE, it can be inferred that it would be rapidly available for permeation across physiologic membranes and, in consequence, it would not present bioavailability concerns. All DE results were higher than 75%, with the exception of product F (Table 2). Statistically significant differences in DE were noted for product F when compared to the reference product and between multisource products (i.e., product F vs. A, C, E, and G; product C vs. B, D, F, and H).

Dissolution results may be influenced by different factors, e.g., disintegration rate, excipient composition, and manufacturing method (*15*). As already mentioned, the lower dissolution results obtained for product F could be related to its higher hardness and disintegration time values; however, no direct relationship was observed between excipient composition and dissolution performance (Table 3). The manufacturing method is not publicly available, so it was

not possible to evaluate this factor.

Table 3. Qualitative Composition of Excipients in Sildenafil (50 mg) Tablets

Excipients		Products							
		A	B	C[b]	D[c]	E (Ref)[b]	F[b]	G	H[b]
Fillers and Diluents	Lactose	+	–	–	+	–	+	+	–
	Microcrystalline cellulose[a]	+	+	+	+	+	+	+	+
	Anhydrous dibasic calcium phosphate	+	+	+	–	+	+	+	+
	Talc[a]	+	–	+	–	–	–	–	+
Disintegrants	Croscarmellose sodium	+	+	+	+	+	+	+	+
Glidants	Colloidal silicon dioxide	–	–	+	+	–	–	–	–
Lubricants	Magnesium stearate	+	+	+	+	+	+	+	+
	Poloxamer[a]	–	–	–	+	–	–	–	–
	Polyethylene glycol	+	+	+	–	–	+	–	+
Binders	Hydroxypropyl methylcellulose[a]	+	–	+	–	–	+	–	–
Coating and coloring agents	Aluminum lacquer	+	+	+	+	–	+	+	+
	Opadry	–	+	–	–	+	–	+	–
	Polyvinyl alcohol[a]	–	–	–	–	–	–	–	+
	Titanium dioxide[a]	+	+	+	–	–	+	–	+
	Coloring agents	+	+	+	+	–	+	+	+
	Triacetin	–	–	–	–	–	+	–	–
Others	Castor oil	+	–	–	–	–	–	–	–
	Simethicone	–	–	–	+	–	+	+	+

[a]*This excipient has multiple functions.*
[b]*Label and/or leaflet inform qualitative and quantitative composition of excipients.*
[c]*Excipient composition was informed only in the label (no leaflet)*
Ref: reference formulation; symbols + and – indicate presence and absence, respectively.

CONCLUSION

Since sildenafil citrate is a BCS class II drug, a waiver of bioequivalence (biowaiver) is not applicable for immediate-release solid oral dosage forms. The Argentine pharmaceutical market is mainly comprised of multisource products. In the particular case of sildenafil formulations, patients can obtain them without prescription and interchange among different brand names with no control. In this scenario, the evaluation of pharmaceutical equivalence becomes an important initial step in the assessment of critical quality attributes of multisource products. Based on the obtained results, it is possible to conclude that the evaluated 50-mg sildenafil immediate-release tablets (reference and multisource products in the Argentine market) can be considered pharmaceutical equivalents. Furthermore, health advice is mandatory during dispensing of these medicinal products.

FUNDING

The authors thank Universidad Nacional del Sur for financial support of this study (PGI24/ZB70).

CONFLICT OF INTEREST

The authors disclosed no conflicts of interest related to this article.

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