# Quality control and comparative dissolution profile of Ibuprofen oral suspension

Controle de qualidade e perfil de dissolução comparativo de ibuprofeno em suspensão oral

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**ABSTRACT**

Due to large number of generic and similar medications containing ibuprofen oral suspension available in the Brazilian market and the need to ensure the quality of these medications, tests to verify the quality and dissolution profile *in vitro* were performed with samples called R (reference), G1, G2 (generic), S1 and S2 (similar), purchased locally. All the products were approved in drug identification, pH, dissolution and quantification assays. In the volume control test, products R and G1 were approved, while products G2, S1 and S2 showed an average volume lower than claimed. Products R, G2 and S1 met the drip test criteria, but products G1 and S2 showed an average content of ibuprofen drop of 71.19% and 66.16% respectively. After evaluation the comparative dissolution profile between products differs as to speed and amount of dissolved drug, and may thus compromise future *in vivo-in vitro* correlations for these formulations.


**KEYWORDS**: Anti-inflammatory, Generic drugs, Similar drugs, Pharmaceutical equivalence.

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**RESUMO**

Devido ao grande número de medicamentos genéricos e similares contendo ibuprofeno suspensão oral disponíveis no mercado brasileiro e a necessidade de assegurar a qualidade desses medicamentos, testes para o controle de qualidade e o perfil de dissolução *in vitro* foram realizados com amostras denominadas R (referência), G1 e G2 (genéricos), S1 e S2 (similares), adquiridas no comércio local. Todos os produtos analisados foram aprovados nos testes de identificação, pH, dissolução e teor do fármaco. No teste de controle de volume, os produtos R e G1 foram aprovados e os produtos G2, S1 e S2 apresentaram volume médio inferior ao declarado. Os produtos R, G2 e S1 cumpriram os critérios do teste de gotejamento, porém os produtos G1 e S2 apresentaram teor médio de ibuprofeno por gota de 71,19% e 66,16% respectivamente. Após a avaliação do perfil de dissolução comparativo entre os produtos, estes apresentaram diferenças na velocidade e quantidade de fármaco dissolvido, podendo assim comprometer futuras correlações *in vivo – in vitro* para estas formulações.

**PALAVRAS-CHAVE**: Anti-inflamatório, Medicamentos genéricos, Medicamentos Similares, Equivalência farmacêutica.

## INTRODUCTION

Inflammation is characterized as an organism response to cell lesion, and it is a complex, dynamic phenomenon promoted by different harmful agents, such as the physical (radiation, Burns, trauma), biological (microorganism, immune reactions) or chemical (caustic substance) type (Silva, 2010; Zaldivar *et al*. 2006).

Ibuprofen (Figure 1) is a non-steroidal anti-inflammatory drug (NSAID) derived from propionic acid, with analgesic, antipyretic, anti-inflammatory, anti-arthritis and antidysmenorrheic action (Korolkovas, 2011).
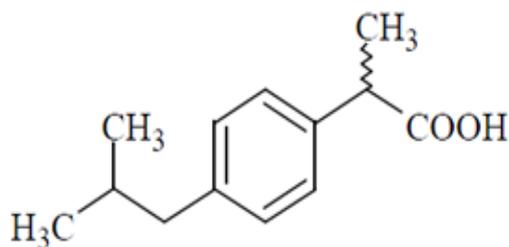


Figure 1. Chemical structure for ibuprofen (The United States Pharmacopeia, 2012).

It is indicated for the treatment of mild to moderate pains, fever, arthritis, non-rheumatic inflammation and prophylaxis of vascular headache. It has been widely used to combat a temperature in pediatric patients, because of its efficacy and proven safety (Farré *et al*. 2005; Kellstein *et al*. 1999). According to Ansel (2007), the liquid pharmaceutical forms are preferred by many patients because they are easy to swallow, the dose is flexible at the time of administration, and it is considered appropriate for elderly patients, children and breastfeeding women. The suspensions are liquid pharmaceutical forms which present solid particles dispersed in an aqueous medium in which these particles are not soluble (Brazilian Pharmacopeia, 2010). They should present appropriate characteristics such as stability, slow sedimentation, easy dispersion after gentle shaking of the container and quick, uniform outflow (Ansel, 2007; Lachman, Liberman & Kanig, 2001).

The interchangeability of the innovative medication with its respective generic is based on the concept of therapeutic equivalence among them. This is verified by proving the pharmaceutical equivalence and bioequivalence (Shargel & Yu, 1999).

The study of dissolution is the most appropriate method for an *in vitro* evaluation of the kinetics of the release of a drug contained in a solid pharmaceutical form and/or in a dispersed/suspended pharmaceutical form, where the cumulative amount of drug which passes into the solution is quantified based on time, and is thus considered an important tool to evaluate the factors that influence bioavailability (Aulton, 2005).

For class II drugs such as ibuprofen with high permeability and low solubility, the dissolution rate is one of the main limiting factors of oral absorption. Thus is it considered essential to establish a strong correlation between the results observed in the *in vitro* dissolution assay and the *in vivo* absorption rate (Dressman *et al*., 1998; Dressman & Fleisher, 1986).

Due to the large number of generic and similar medications containing an oral suspension of ibuprofen available in the Brazilian market and the need to ensure the quality of these medications, the purpose of this study was to evaluate quality control and *in vitro* dissolution profile of the reference medication, two generic medications and two similar ones.

## MATERIAL AND METHODS

### Samples, Chemical Reference Substance (CRS) and reagents

Ibuprofen Chemical Reference Substance (CRS) - Lote K0J008 - The United States Pharmacopeia. Oral suspensions of ibuprofen 50 mg/ml purchased in local businesses were used. They were classified as similar (S1 and S2), generic (G1 and G2) and reference medication (R). The other reagents were all analytic grade.

### Equipment

The development and validation of the assay was performed on a Shimadzu® LC system (Kyoto, Japan), with an LC-10AD pump, detector with variable wave length UV/Vis model SPD-10Avp, controller SLC-10Dvp, automatic integrator with Class VP, automatic injector SIL-10-Avp and column oven CTO-10Asvp. Phenomenex® Luna $C_8$ Column , 150 x 4,6 mm, the size of particle 5 μm. Dissolutor, PTWS Pharmatest®, Viscosimeter, Brookfield®, and Ultrabasic Potentiometer, Denver®.

## EVALUATION OF QUALITY CONTROL

### Identification

The samples were identified by thin layer chromatography (TLC) (The United States Pharmacopeia, 2012).

### Determination of the pH

The pH was determined directly in the samples, with the help of a previously calibrated potentiometer (The United States Pharmacopeia, 2012).

### Viscosity

Apparent viscosity was determined directly in the samples, at a temperature of 25 ºC, taking 1 minute to adjust the velocity factor (Brasil, 2010a). For samples R, G2 and S1 spindle 3 was used, and for G1 and S2 spindle 2, all at a velocity of 100 rpm.

### Determination of volume

Ten flasks chosen randomly were weighed individually, washed first with water and then ethanol, dried at room temperature and weighed again. The difference between the two weighings was the weight of the content (Brasil, 2010a).

### Drip test

The drip test was evaluated using 10 units of each product at a controlled temperature of 20 °C, where the mass relative to the number of drops corresponding to 1 mL was determined, as declared by the manufacturer, and the amount of drug per drop for each unit tested (Brasil, 2010a).

**Determination of density**

The density was determined using a pycnometer previously calibrated with ultra-purified water and samples at a temperature of 20 °C (Brasil, 2010a).

**Dosing**

The ibuprofen content in the suspensions was determined using the following chromatographic conditions: column $C_8$ (4.6 mm x 15 cm, 5 µm), mobile phase phosphoric acid 0.01 *M* and acetonitrile (63:37, v/v), flow of 2 mL/minute, the injection volume was 5 µl, PDA detection at 220 nm (The United States Pharmacopeia, 2012).

**Preparation of the standard curve for dosing**

About 160 mg of internal standard benzophenone were weighed for a 50 mL balloon in order to obtain a solution at 3.2 mg/mL. Then precisely 50 mg of ibuprofen CRS were weighed for a 50 mL volumetric balloon; 30 mL of diluent solution were added, composed of acetonitrile and water (1:1). This was taken to the ultrasound for 15 minutes and the volume was completed with the same diluent (1000 µg/mL). The standard curve was prepared from this solution with concentrations ranging from 400 µg/mL to 600 µg/mL. The procedure was performed in triplicate, and the solutions obtained were submitted to analysis by HPLC using the previously described chromatographic conditions.

**Preparation of the samples for dosing**

A pool was prepared with 10 flasks of each product. Using this pool the mass equivalent to 60 mg of ibuprofen was measured; 30 mL of diluent were added for a 50 mL volumetric balloon volumetric balloon. It was placed for 15 minutes in ultrasound and the volume was completed with diluent. A 20 mL aliquot was removed and transferred to a 50 mL volumetric balloon, 1 mL of the internal standard benzophenone solution was added and the volume was

completed with acetonitrile, homogenized and filtered into vials. The samples were prepared in triplicate and submitted to analysis by HPLC using the previously described chromatographic conditions.

**Dissolution and dissolution profile**

The parameters described in Table 1 were used to perform dissolution, and the collection was performed within a 60 minute period (The United States Pharmacopeia, 2012). The same conditions of the dissolution test were used to analyze the dissolution profile, with collections of 10 mL used at 5, 10, 15, 20 and 40 minutes. The volume removed was replaced with the same dissolution medium.

Table 1. Parameters utilized for realization of dissolution test.

| Parameter | Condition |
|---|---|
| Dissolution medium and temperature | Buffer solution pH 7.2/37.0 ± 0.5 ℃ |
| Volume of dissolution médium | 900 mL |
| Apparatus/speed of rotation | Paddle/50 rpm |
| Volume of sample utilized | 10 mL (equivalent of 500 mg of ibuprofen) |
| Concentration of sample | 555,55 µg/mL |

**Preparation of the standard curve for dissolution and dissolution profile**

Precisely about 50 mg of ibuprofen CRS were weighed for a 50 mL volumetric balloon, 30 mL of diluent solution composed of acetonitrile and water (1:1) were added. The solution was taken to the ultrasound for 15 minutes and the volume was completed with the same solvent (1000 µg/mL). Based on this stock solution, a standard curve was prepared with concentrations from 110 µg/mL to 650 µg/mL. The procedure was performed in triplicate and the solutions obtained were analyzed using the same parameters for analysis described for dosing.

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**Preparation of the samples for dissolution and dissolution profile**

A pool of 10 flasks of each product was prepared. Syringes were filled with a quantity equivalent to 500 mg of ibuprofen (approximately 10 mL) while maintaining constant shaking. The samples were released slowly and constantly into the vessels while the apparatus was moving. The syringes were weighed before and after the addition of the samples and the difference in weight was related to density in order to calculate the real quantity added to each vessel. The amount of dissolved ibuprofen was determined by a chromatographic method using the same conditions as for the dosing assay.

**RESULTS AND DISCUSSION**

**Identification**

The Rf values of standard and medications were similarity, as well as the intensity and form of spot obtained with the products. The medications analyzed complied with the identification test (Figure 2) to which they were submitted.
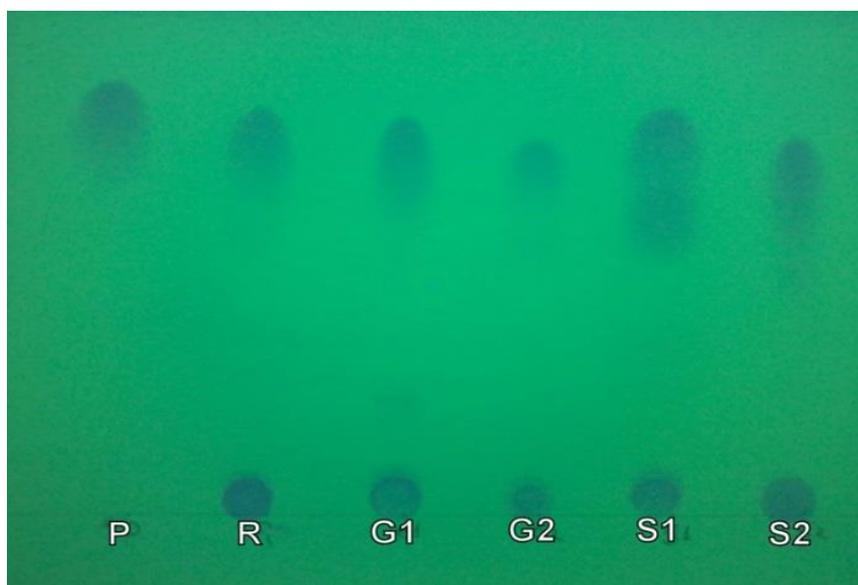


Figure 2. Thin layer chromatography (TLC) of samples and CRS of ibuprofen (P = CRS of ibuprofen; R = reference medication; G1 = Generic medication 1; G2 = Generic medication 2; S1 = Similar medication 1 and S2 = Similar medication 2).

## Determination of the pH

The values obtained determining pH (Table 2) in the samples analyzed, are in accordance with the product monograph which allows a variation range of 3.6 to 4.6.

## Density and Viscosity

The results obtained in determining the density and apparent viscosities are shown in Table 2.

According to resolution – RDC Nº 31 of August 11, 2010 (Brasil, 2010), the density and viscosity assays do not represent parameters that approve or reject the product within the scope of studies of pharmaceutical equivalence, rather they are considered informative assays. The difference in relation to the density and viscosity values obtained for the products are generally associated with the characteristics such as stability and flow properties of each formulation (Aulton, 2005).

## Volume determination

The volume determination test is needed for liquid products in containers for multiple doses and liquid products in single dose containers. According to the Brasil (2010a) the mean volume of the units analyzed should not be less than the volume declared by the manufacturer (30 mL) and no individual volume of the units tested is less than 95.0% (28.5 mL) of the declared volume. None of the samples analyzed presented an individual volume of less than 95% of the declared volume, but the mean volume of samples G2, S1, S2 was less than the declared volume (Table 2) and thus these samples were rejected.

Table 2. Results obtained of determination of pH, density, viscosity and determination of volume of ibuprofen oral suspension.

| Samples | pH | Density (g/mL) | Viscosity* (cP) | Average volume (mL) |
|---------|------|------|-------|-------|
| R | 4,01 | 1,12 | 176,0 | 31,37 |
| G1 | 4,36 | 1,05 | 144,8 | 30,02 |
| G2 | 3,86 | 1,09 | 229,0 | 29,39 |
| S1 | 4,44 | 1,22 | 510,0 | 29,42 |
| S2 | 4,08 | 1,16 | 80,8 | 29,56 |

*average values of two determination. cP - centipoise

**Drip test**

All manufacturers declared that 10 drops correspond to 1 mL of the product that is being studied. In order for the products to comply with the test requirements to determine the number of drops and drug content per drop, the difference allowed in relation to the number of drops per 1 milliliter of the tested medication is up to ±10% in relation to the value declared for the reference medication and the amount of drug per drop should be between 85.0% and 115.0% (Brasil, 2010; Brasil, 2010a). The medications (G2 and S1) complied with the criteria, medication (G1) presented 15 drops per 1 milliliter and a mean content of ibuprofen per drop of 71.22%, while medication (S2) presented 16 drops per 1 milliliter and a mean drug content of 66.19%. The mean results are shown in Table 3.

Table 3. Results found for drip test for samples.

| Sample | Number of drop* | Quantity of drug per drop* (mg/drop) | Quantity declared* (%) | RSD**(%) |
|--------|--------|--------|--------|--------|
| R | 10,45 | 5,08 | 101,50 | 3,38 |
| G1 | 14,94 | 3,56 | 71,19 | 2,26 |
| G2 | 10,45 | 5,18 | 103,59 | 1,82 |
| S1 | 10,68 | 4,95 | 98,90 | 2,40 |
| S2 | 15,95 | 3,31 | 66,16 | 1,95 |

*average values obtained with ten samples; **relative standard deviation

The drop size is influenced by several factors, such as density of the liquid, temperature, surface tension, dropper design, diameter of the opening at the tip of the dropper, and flask elasticity (Ferreira, 2007). If these factors are not correctly evaluated, they may interfere directly in the dosage administered and, consequently in the treatment of the patient.

**Quantitative analysis**

The United States Pharmacopeia (2012) specifies that the ibuprofen suspension should contain at least 90.0% and at most 110.0% of the declared amount of active ingredient. The results obtained in dosing the products were interpolated in the previously constructed analytic curve (y = 6325.7x + 71401), ensuring a linearity in the range of 400 to 600 μg/mL ($r^2$=0.9995), and did not present a deviation of linearity (*p=0.05*), when evaluated by

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Analysis of Variance (ANOVA). All the samples were in accordance with the values specified, as described in Table 4.

Table 4. Results obtained of quantitative analysis of oral suspension.

| Samples | Individual quantities (%) | Average of quantitie (%) | RSD[*] (%) |
|---------|---------------------------|--------------------------|----------|
| R | 106,70<br>105,35<br>106,03 | 106,03 | 0,64 |
| G1 | 105,55<br>107,89<br>105,70 | 106,38 | 1,23 |
| G2 | 108,99<br>108,91<br>106,79 | 108,23 | 1,15 |
| S1 | 107,27<br>104,75<br>104,79 | 105,60 | 1,37 |
| S2 | 105,46<br>106,31<br>104,91 | 105,56 | 0,67 |

[*]relative standard deviation

**Dissolution and dissolution profile *in vitro***

Dissolution tests are a very important tool to evaluate the different medication production stages, making it possible to monitor the stability of formulations, as well as possible deviations from quality (Dressman *et al*., 1998).

The results obtained in the dissolution tests and in the dissolution profiles were interpolated in the previously constructed analytic curve (y = 5357.3x + 6699.8), ensuring linearity in the 110 to 650 μg/m range ($r^2$=0.999), and not presenting a deviation of linearity (*p=0.05*) when evaluated by Analysis of Variance (ANOVA). The results of the dissolution test are described in Table 5 and those of the dissolution profiles in Table 6 and are represented graphically in Figure 3.

Table 5. Results obtained of dissolutions tests of oral suspensions containing ibuprofen.

| Samples | Larger quantity dissolved (%) | Smaller amount dissolved (%) | Average dissolved (%) | RSD[*](%) |
|---------|------------------------------|------------------------------|------------------------|-----------|
| R | 91,32 | 85,98 | 88,63 | 2,53 |
| G1 | 95,22 | 91,04 | 92,96 | 1,53 |
| G2 | 86,69 | 85,58 | 86,13 | 1,92 |
| S1 | 104,39 | 98,69 | 101,09 | 2,42 |
| S2 | 104,48 | 99,87 | 102,41 | 1,79 |

[*]relative standard deviation; n=6

Tabela 6. Resultados médios obtidos no estudo do perfil de dissolução do medicamento referência, dos genéricos G1 e G2 e dos similares S1 e S2.

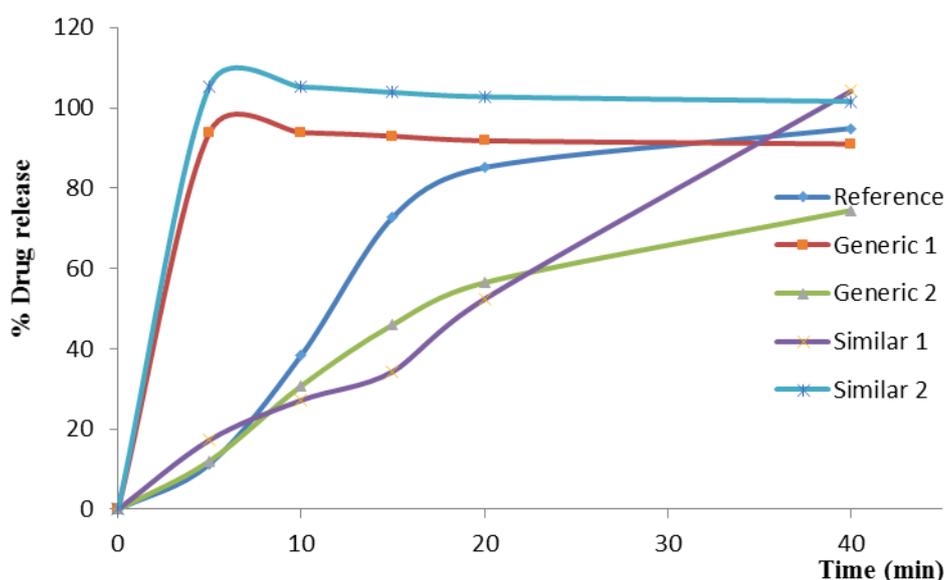| | % Dissolved | | | | |
|-------------|-------|-------|-------|-------|--------|
| Tempo (min) | R | G1 | G2 | S1 | S2 |
| 0 | 0 | 0 | 0 | 0 | 0 |
| 5 | 11,33 | 93,92 | 12,05 | 17,26 | 105,41 |
| 10 | 38,35 | 93,86 | 30,74 | 27,15 | 105,23 |
| 15 | 72,76 | 92,96 | 45,99 | 34,32 | 103,85 |
| 20 | 85,13 | 91,83 | 56,46 | 52,37 | 102,77 |
| 40 | 94,88 | 90,99 | 74,46 | 104,16 | 101,59 |



Figure 3. Dissolution profiles of samples containing ibuprofen.

The products evaluated complied with the approval criteria for the dissolution test and this was that no less than 80.0% (Q) of the declared quantity of ibuprofen dissolved in 60 minutes.

The dissolution profiles of medications presented were different from each other, especially as regards velocity and percentage of the drug released. Medications G1 and S2 released more than 85.0% of the drug already during the first 5 minutes of the test, and were classified as very quick release (Brasil, 2010). The reference medications and S1 released 94.88% and 104.16% in 40 minutes, but during this same time formulation G2 released 74.46%.

In order to compare the dissolution profiles the difference ($f_1$) and similarity ($f_2$) factors were calculated between reference product R and similar product S1, since product S1 presented a type of dissolution corresponding to product R. The profiles studied are considered similar when the comparison between them results in values of $f_1$ between 0 and 15 and $f_2$ between 50 and 100 (Brasil, 2010). After the test the results obtained for ($f_1$) and ($f_2$) were 32.27 and 31.29 respectively, showing non similarity between the profiles.

The formulations evaluated showed a great difference when compared to the reference product. The comparison (reference x G2) could not be performed because product G2 and reference do not release simultaneously 85.0% of the drug at least at one point of collection, since this is one of the criteria for the use of the factors $f_1$ and $f_2$ as parameters for comparison. All profiles presented a RSD of less than 20% in the first 15 minutes and less than 10% at the other times, complying with the determinations of RDC 31 (Brasil, 2010).

## CONCLUSION

The results obtained after the quality control of the products evaluated showed that all products complied with the identification tests and pH, showing viscosity and density values appropriate to the characteristics of each formulation. In the volume control test products G2, S1, S2, did not comply with the test. After the evaluation of the drip test, products R, G2 and S1 complied with the test criteria. All the products complied with the dosing and dissolution test but presented a great difference in relation to velocity and quantity of drug released, compared to the reference product, and thus may compromise future tests on the comparative dissolution profile between formulations.

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