dx.doi.org/10.14227/DT320125P42

Hypothetical Plasma Concentration-Time Profiles of Amiodarone-HCl From In Vitro Release Data

José Raúl Medina-López[1*] and Felipe Dino Reyes-Ramírez[1]
[1]*Departamento Sistemas Biológicos, Metropolitan Automomous University-Xochimilco, Mexico City, Mexico.*

e-mail: rmlopez@correo.xoc.uam.mx

ABSTRACT

Introduction: This study aimed to determine the hypothetical plasma concentrations of amiodarone-HCl (AMD) formulations from in vitro release data. **Methods**: The dissolution method used a United States Pharmacopeia paddle apparatus at 75 rpm. The dissolution medium was 0.1 N hydrochloric acid (pH 1.2) with 1% sodium dodecyl sulfate (SDS), pH 4.5 acetate buffer with 1% SDS, and pH 6.8 phosphate buffer with 1% SDS. The reference and a generic tablet formulation (200 mg) were studied. Dissolution parameters were compared and analyzed for statistically significant differences (e.g., mean dissolution time, cumulative drug release, dissolution efficiency, and others). Hypothetical plasma concentration-time profiles were calculated with dissolution data and reported pharmacokinetic information for AMD using a convolution approach. **Results**: Similarity factor (f_2) analysis indicated that all dissolution profiles were similar; however, some statistically significant differences ($p < 0.05$) were noted in the dissolution parameters. According to established criteria of $R^2_{adjusted}$ and AIC values, AMD dissolution behavior was best explained by the Weibull model. **Conclusion**: Dissolution data for the reference formulation at pH 4.5 gave an acceptable prediction error for C_{max} and $AUC_{0\text{-}inf}$.

KEYWORDS: Amiodarone-HCl (AMD), plasma concentrations, prediction error, dissolution

INTRODUCTION

Amiodarone-HCl (AMD) tablets are suggested for the treatment of life-threating ventricular and supraventricular arrhythmias and atrial fibrillation (*1*, *2*). AMD is a compound with low solubility (0.35 mg/mL), high permeability, high protein binding (> 96%), and moderate bioavailability (35–65%) (*1*). Food promotes drug absorption, which improves bioavailability (*3*). AMD is classified by the Biopharmaceutic Classification System (BCS) as a class 2 drug (*2*, *4*).

Dissolution studies are an important tool to establish the release performance of poorly soluble drugs. Compendial quality control tests for AMD tablets are performed using United States Pharmacopeia (USP) apparatus 2 (paddle) at an agitation rate of 100 rpm with 1000 mL of 1% sodium dodecyl sulfate (SDS); and no less than 80% of the dose should dissolve in 60 min (*5*).

In vitro release of AMD formulations has been studied by some authors. For context, in Brazil, Rubim et al reported increased solubility and dissolution of AMD with a solid dispersion technique using an inclusion complex containing a hydrophilic carrier (dissolution media were distilled water and aqueous solution of pH 1.2, 4.5, and 6.8 in USP apparatus 2 at 50 rpm for 90 min) (*6*). In Australia, Ngo et al compared different commercially available AMD tablets using a high-performance liquid chromatography (HPLC) method (USP apparatus 2 at 100 rpm for 90 min with 1000 mL 1% SDS in water); however, only the innovator and two out of three generic (multisource) drug

*Corresponding author

products achieved the pharmacopeial criterion (Q ≥ 80% in 30 min) (*4*).

Considering the variability observed in bioequivalence studies, the narrow therapeutic ratio, and diversity in dissolution results, the objective of the present study was to determine the release behavior of AMD tablets available to the Mexican population, considering biowaiver conditions, and to estimate the hypothetical plasma concentration-time profiles (*3*, *7*).

METHODS

AMD tablets of the reference formulation (Cordarone 200 mg, Sanofi-Aventis de México, S.A. de C.V. Ocoyoacac, Mexico; lot no. BMXA002; expiration October 2023) and a generic formulation (Braxan 200 mg, Armstrong Laboratorios de México, S.A. de C.V. Mexico City, Mexico; lot no. 21080057; expiration Oct 2023) were tested. HCl, methanol, acetic acid, sodium acetate, sodium hydroxide pellets, and sodium phosphate monobasic crystals were supplied by J.T.Baker-Mexico. AMD reference substance was provided by Sigma-Aldrich Co. (St. Louis, MO, USA).

Dissolution Test

Dissolution of AMD tablets was tested under biowaiver conditions using a paddle apparatus (Sotax, AT7-Smart) at 75 rpm with 900 mL of 0.1 N hydrochloric acid (pH 1.2) and 1% SDS, pH 4.5 acetate buffer with 1% SDS, and pH 6.8 phosphate buffer with 1% SDS at 37.0 ± 0.5 °C. Automatic samples were taken every 5 min over 60 min (n = 12) and filtered with glass fiber prefilters (Millipore, Ireland). Dissolution medium was recirculated after each sample was taken. AMD was identified spectrophotometrically at 243 nm (Lambda 25, PerkinElmer, USA) with the support of solutions with known concentrations ranging from 1.87–30 µg/mL.

Data Analysis

To compare AMD dissolution curves for the reference and generic product, the difference (f_1) and similarity (f_2) factors at each pH level were calculated. Acceptable f_1 values are 0–15, and f_2 values are 50 – 100 (*8*).

The cumulative amount of drug released at the last sampling time (Q_{60}), mean dissolution time (MDT), and dissolution efficiency (DE) were computed and compared using the Student's t-test. AMD release behavior was studied with following mathematical models: Makoid-Banakar, Weibull, Korsmeyer-Peppas, Peppas-Sahlin, and logistic. The adjusted determination coefficient ($R^2_{adjusted}$) and Akaike Information Criterion (AIC) were used to determine the best fitting model (*9*).

The f_1, f_2, DE, and MDT values were computed by the Excel add-in DDSolver (*10*). Time to dissolve 50% of dose ($t_{50\%}$) was calculated with a hyperbola model.

Simulation of Plasma Concentrations

AMD plasma levels were calculated with a convolution method as follows (*11*). Pharmacokinetic parameters were used to build drug levels in function of time, including elimination rate (k_e), bioavailability factor (F), and volume of distribution (V_d) (*12*, *13*). Peak plasmatic level (C_{max}) and area under the plasma concentration-time curve from zero time to infinity ($AUC_{0\text{-}inf}$) were computed using the Excel add-in PKSolver (*14*).

Data for the reference AMD formulation in humans were then used to calculate prediction error (%PE; < 10% is optimal) for pharmacokinetic parameters C_{max} and $AUC_{0\text{-}inf}$ according to the following equation (*3*, *15*):

$$\%\mathrm{PE} = \frac{(observed\ value - predicted\ value)}{observed\ value} \times 100.$$

RESULTS AND DISCUSSION

Dissolution profiles for reference and generic AMD formulations are shown in Figure 1. Despite addition of SDS in all dissolution media, less than 50% was released from both formulations at pH 6.8 after 60 min. As shown in Table 1, both formulations had similar dissolution profiles based on f_2 values. Statistical differences (p < 0.05) in Q_{60} and DE values were noted at all pH levels, but MDT significant differences were only found at pH 1.2. Comparison of dissolution profiles by DE and MDT was made to facilitate results comparison; the differences had no physiological meaning (*16*).

These findings agree with results reported by others. Awan et al studied dissolution profiles of AMD powder, a commercial product (Cordarone 200 mg), and a test formulation (AMD nanocrystals equivalent to 200 mg) using the paddle apparatus at 100 rpm with 900 mL of distilled water and pH 1.2 and pH 6.8 buffer solutions. In all dissolution media, less than 60% of the AMD marketed product dissolved after 90 min (*17*).

Wang et al carried out an in vitro study of commercial tablets (Cordarone 200 mg) and AMD inside an inclusion complex using the paddle apparatus at 50 rpm with 900 mL of distilled water, pH 1.0 HCl, and pH 4.5 and 6.8 buffer solutions. After 90 min with the commercial formulation, limited release (< 10%) was observed at pH 1.0 and 6.8 ,

and less than 50% was released with distilled water; at pH 4.5, 84.2% of dissolved drug was released (*18*).

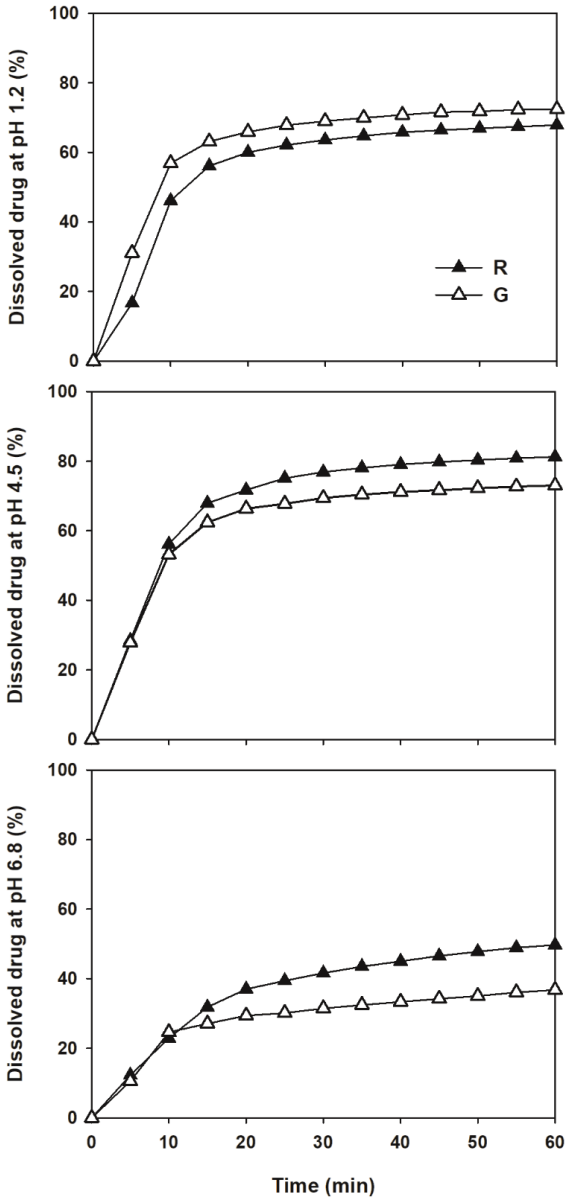

Figure 1. Dissolution profiles (mean values, n = 12) of reference and generic AMD tablet formulations using the paddle apparatus at 75 rpm with 900 mL of 0.1 N HCl (pH 1.2) + 1% SDS; acetate buffer + 1% SDS (pH 4.5); and phosphate buffer + 1% SDS (pH 6.8).
AMD: amiodarone-HCl; G: generic; HCl: hydrochloric acid; R: reference; SDS: sodium dodecyl sulfate.

Kinetic modeling of in vitro drug release for the reference and generic AMD formulations is shown in Table 2. According to established criteria of $R^2_{adjusted}$ and AIC values, AMD dissolution behavior was best explained by the Weibull model, which emphasizes the S-shaped profile (*19*). The $R^2_{adjusted}$ and AIC values were selected due observed differences in other parameters (*20*). The model-dependent parameter $t_{50\%}$ (derived from adjustment to hyperbola model) and Td (derived from Weibull function) are listed in Table 1; statistical differences ($p < 0.05$) were noted for $t_{50\%}$ but not Td. Because a low Q_{60} value (< 50%) was observed for AMD at pH 6.8, the $t_{50\%}$ and Td values were not calculated.

After applying the convolution methodology and adjusting the predicted AMD plasma levels to a compartment model, the main pharmacokinetic parameter values were calculated. The hypothetical C_{max} and $AUC_{0\text{-}inf}$ were associated with human data to calculate prediction error (*3*). As shown in Table 3, prediction error was less than 10% for both parameters of the reference formulation only at pH 4.5. Therefore, using the paddle apparatus at 75 rpm with pH 4.5 acetate buffer and 1% SDS was suitable to predict AMD plasma concentrations similar to those reported in an in vivo study. The predicted dissolution profiles obtained with in vitro release data at pH 4.5 are shown in Figure 2. The convolution method was validated, as PE < 10% was found (*15*).

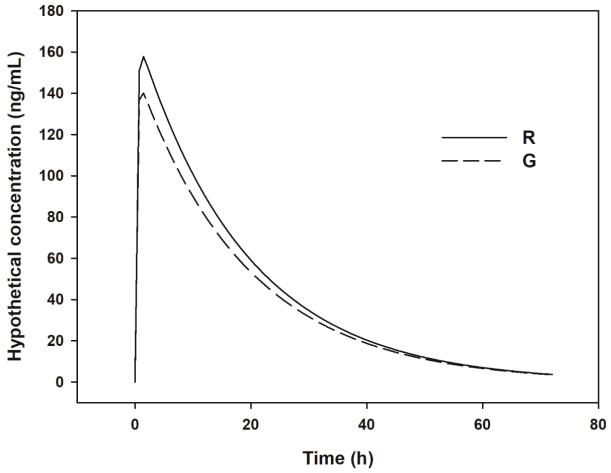

Figure 2. Hypothetical plasma concentration-time profiles of reference and generic AMD tablet formulations calculated with dissolution data at pH 4.5.
AMD: amiodarone-HCl; G: generic; HCl: hydrochloric acid; R: reference; SDS: sodium dodecyl sulfate.

It is preferable to establish an in vitro-in vivo correlation (IVIVC) for formulations with absorption limited by the dissolution rate (*21*). Two studies of IVIVC using AMD tablets have been reported (*2, 22*). The first was a recompilation of three bioequivalence studies of a reference and three commercial generic products using the paddle apparatus at 75 rpm with 0.1 M pH 5.0 acetate buffer and 1% SDS for 120 min; a poor level B correlation was found ($p = 0.033$) (*2*). The authors concluded that bioequivalence studies should be carried out to ensure the interchangeability of AMD multi-source formulations (*2*). The second IVIVC study estimated

Table 1. Dissolution Parameters of Reference and Generic AMD Tablet Formulations

pH[a]	Product	f_1	f_2	Q_{60} (%)[b]	DE (%)[b]	MDT (min) [b]	$t_{50\%}$ (min) [b]	Td (min) [b]
1.2	R	-	-	67.85 ± 0.89	55.80 ± 0.68	10.62 ± 0.36	14.70 ± 0.55	17.41 ± 7.25
	G	11.20	57.91	72.42 ± 0.29*	62.18 ± 0.58*	8.48 ± 0.44*	9.15 ± 0.59*	8.60 ± 1.12
4.5	R	-	-	81.25 ± 0.68	67.90 ± 0.77	9.85 ± 0.37	8.55 ± 0.37	9.23 ± 0.34
	G	9.05	58.83	73.03 ± 0.78*	61.79 ± 0.72*	9.23 ± 0.25	10.33 ± 0.54*	8.64 ± 0.31
6.8	R	-	-	49.67 ± 0.05	36.78 ± 0.54	15.44 ± 0.72	†	†
	G	23.34	50.94	36.73 ± 0.47*	28.54 ± 0.46*	13.38 ± 0.36	†	†

[a]Dissolution media: 0.1 N HCl (pH 1.2) + 1% SDS; acetate buffer + 1% SDS (pH 4.5); phosphate buffer + 1% SDS (pH 6.8).
[b]Values are expressed as mean ± standard error (n = 12).
**p < 0.05; †Less than 50% of dissolved drug was found at 60 min, so no $t_{50\%}$ and Td data were calculated.*
AMD: amiodarone-HCl; DE: dissolution efficiency; f_1: dissimilarity factor; f_2: similarity factor; G: generic; HCl: hydrochloric acid; MDT: mean dissolution time; R: reference, SDS: sodium dodecyl sulfate; Td: derived from the Weibull model.

Table 2. Kinetic Release Parameters of Reference and Generic AMD Tablet Formulations (n = 12).

pH[a]	Parameter	Product	Makoid-Banakar	Peppas-Sahlin	Korsmeyer-Peppas	Logistic	Weibull
1.2	$R^2_{adjusted}$	R	0.8941	0.8709	0.7268	0.8226	0.9977
		G	0.8986	0.8901	0.7350	0.8289	0.9974
	AIC	R	68.94	71.40	79.97	74.32	17.38
		G	62.83	63.78	73.79	67.77	17.88
4.5	$R^2_{adjusted}$	R	0.9270	0.9143	0.7692	0.8226	0.9953
		G	0.9142	0.9050	0.7521	0.8559	0.9954
	AIC	R	65.06	67.32	79.16	74.32	33.24
		G	63.42	65.22	76.12	69.44	16.01
6.8	$R^2_{adjusted}$	R	0.9756	0.9700	0.9412	0.9096	0.9901
		G	0.9017	0.8978	0.8455	0.8667	0.9906
	AIC	R	42.86	45.20	58.97	51.46	30.95
		G	51.19	51.66	55.95	54.08	22.27

[a]Dissolution media: 0.1 N HCl (pH 1.2) + 1% SDS; acetate buffer + 1% SDS (pH 4.5); phosphate buffer + 1% SDS (pH 6.8).
AIC: Akaike Information Criterion; AMD: amiodarone-HCl; G: generic; HCl: hydrochloric acid; R: reference; SDS: sodium dodecyl sulfate.

AMD in vivo release from the pharmacokinetics of desethylamiodarone (active metabolite) using reference tablets (200 mg) and a test formulation and the paddle apparatus at 100 rpm with 1000 mL of SDS (10 mg/mL) in ultrapure water (*22*). After 60 min, both formulations released the complete dose. The authors proposed the correlation of in vitro drug release with pharmacokinetics of the active metabolite (*22*).

Given the variability with AMD's in vitro and in vivo performance, more research is needed to establish an IVIVC for AMD tablets. This work simulated AMD plasma levels from dissolution data obtained under biowaiver conditions. The proposed methodology is an option to facilitate biopharmaceutical evaluation of AMD oral dosage forms and ensure interchangeability between different formulations.

Table 3. Prediction Error Parameters of Reference and Generic AMD Tablet Formulations.

Product	Parameter	pH[a]		
		1.2	4.5	6.8
R	C_{max}	21.75%	7.02%	41.06%
	$AUC_{0\text{-}inf}$	20.94%	4.96%	43.31%
G	C_{max}	18.64%	17.14%	57.18%
	$AUC_{0\text{-}inf}$	14.49%	14.21%	57.65%

[a]Dissolution media: 0.1 N HCl (pH 1.2) + 1% SDS; acetate buffer + 1% SDS (pH 4.5); phosphate buffer + 1% SDS (pH 6.8).
AMD: amiodarone-HCl; G: generic; HCl: hydrochloric acid; SDS: sodium dodecyl sulfate; R: reference.

CONCLUSION

The present study proposes hypothetical AMD plasma concentration-time profiles on the basis of in vitro release data for the reference and a generic formulation obtained

under biowaiver conditions. The best conditions to simulate in vivo behavior were obtained with data from the reference formulation using USP apparatus 2 at 75 rpm with 900 mL of pH 4.5 acetate buffer and 1% SDS. At this pH, significant differences in model-independent parameters Q_{60} and DE as well as the model-dependent parameter $t_{50\%}$ reflected differences in the proposed in vivo behavior of the generic formulation. The dissolution conditions and proposed predictions can be used to support better design of AMD formulations.

DISCLOSURES

The authors received no financial support for this work and have no conflicts of interest.

REFERENCES

1. Rodríguez-Fernández, K.; Gras-Colomer, E.; Climente-Martí, M.; Mangas-Sanjuán, V.; Merino-Sanjuán, M. Pharmacometric characterization of entero-hepatic circulation processes of orally administered formulations of amiodarone under complex binding kinetics. *Eur. J. Pharm. Sci*. **2022**, *174*, 106198. DOI: 10.1016/j.ejps.2022.106198
2. Emami, J. Comparative in vitro and in vivo evaluation of three tablet formulations of amiodarone in healthy subjects. *Daru J. Pharm. Sci*. **2010**, *18* (3), 193–199.
3. dos Santos Filho, H. O.; Ilha, J. O.; Silva, L. C.; Borges, A.; Mendes, G. D.; De Nucci, G. Comparative bioavailability study with two amiodarone tablet formulations administered with and without food in healthy subjects. *Arzneimittelforschung* **2007**, *57* (9), 582–590. DOI: 10.1055/s-0031-1296653.
4. Ngo, S. N. T.; Barnes, T. Is there variability in drug release and physical characteristics of amiodarone chloride from different commercially available tablets? Possible therapeutic implications. *Int. J. Pharm. Pract*. **2010**, *18* (4), 245–248. DOI: 10.1111/j.2042-7174.2010.00037.x.
5. Amiodarone Tablets Monograph. In *USP 43-NF 38*. The United States Pharmacopeial Convention, Inc., 2021.
6. Rubim, A. M.; Rubenick, J. B.; Gregolin, E.; Laporta, L. V.; Leitenberg, R.; Rolim, C. M. B. Amiodarone hydrochloride: enhancement of solubility and dissolution rate by solid dispersion technique. *Braz. J. Pharm. Sci*. **2015**, *51* (4), 957–966. DOI: 10.1590/S1984-82502015000400021.
7. Atanasova, I.; Terziivanov, D. Evaluation of average bioequivalence of two oral formulations of amiodarone hydrochloride after single administration to healthy volunteers. *Clin. Drug Investig*. **2001**, *21* (6), 423–428. DOI: 10.2165/00044011-200121060-00005.
8. Moore, J. W.; Flanner, H. H. Mathematical comparison of dissolution profiles. *Pharm. Technol*. **1996**, *20* (6), 64–74.
9. Yuksel, N.; Kanik, A. E.; Baykara, T. Comparison of in vitro dissolution profiles by ANOVA-based, model-dependent and -independent methods. *Int. J. Pharm*. **2000**, *209* (1-2), 57–67. DOI: 10.1016/S0378-5173(00)00554-8.
10. Zhang, Y.; Huo, M.; Zhou, J.; Zou, A.; Li, W.; Yao, C.; Xie, S. DDSolver: an add-in program for modeling and comparison of drug dissolution profiles. *AAPS J*. **2010**, *12* (3), 263–271. DOI: 10.1208/s12248-010-9185-1.
11. Qureshi, S. A. In vitro–in vivo correlation (IVIVC) and determining drug concentrations in blood from dissolution testing – a simple and practical approach. *Open Drug Deliv. J*. **2010**, *4*, 38–47. DOI: 10.2174/1874126601004010038.
12. Mujović, N.; Dobrev, D.; Marinković, M.; Russo, V.; Potpara, T. S. The role of amiodarone in contemporary management of complex cardiac arrhythmias. *Pharmacol. Res*. **2020**, *151*, 104521. DOI: 10.1016/j.phrs.2019.104521.
13. Marrafa, J. M. Amiodarone. In *Encyclopedia of Toxicology*; Wexler, P., Ed.; Academic Press, 2014; pp 197–199. DOI: 10.1016/B978-0-12-386454-3.00691-6
14. Zhang, Y.; Huo, M.; Zhou, J.; Xie, S. PKSolver: An add-in program for pharmacokinetic and pharmacodynamic data analysis in Microsoft Excel. *Comput. Methods Programs Biomed*. **2010**, *99* (3), 306–314. DOI: 10.1016/j.cmpb.2010.01.007.
15. Rastogi, V.; Yadav, P.; Lal, N.; Rastogi, P.; Singh, B. K.; Verma, N.; Verma, A. Mathematical prediction of pharmacokinetic parameters-an in-vitro approach for investigating pharmaceutical products for IVIVC. *Future J. Pharm. Sci*. **2018**, *4* (2), 175–184. DOI: 10.1016/j.fjps.2018.03.001.
16. Adams, E.; Coomans, D.; Smeyers-Verbeke, J.; Massart, D. L. Non-linear mixed effects models for the evaluation of dissolution profiles. *Int. J. Pharm*. **2002**, *240* (1-2), 37–53. DOI: 10.1016/S0378-5173(02)00127-8.
17. Awan, A. M.; Farid, A.; Shah, S. U.; Khan, D.; Ur Rehman, F.; Dar, M. J.; Iftikhar, T.; Ghazanfar, S.; Galanakis, C. M.; Alamri, A. S.; et al. Nanocrystals-mediated oral drug delivery: enhanced bioavailability of amiodarone. *Pharmaceutics* **2022**, *14* (6), 1300. DOI: 10.3390/pharmaceutics14061300.
18. Wang, D.; Chen, G.; Ren, L. Preparation and characterization of the sulfobutylether-β-cyclodextrin inclusion complex of amiodarone hydrochloride with enhanced oral bioavailability in fasted state. *AAPS PharmSciTech* **2017**, *18* (5), 1526–1535. DOI: 10.1208/s12249-016-0646-4.
19. Ilango, K. B.; Kavimani, S. A systematic review of mathematical models of pharmaceutical dosage forms. *Int. J. Curr. Pharm. Rev. Res* **2015**, *6* (1), 59–70.
20. Das, S. K.; Chakraborty, S.; Bose, A.; Rajabalaya, R.; Khanam, J. Effects of the preparation technique on the physicochemical characteristics and dissolution improvement of ketoprofen-SDE7-β-CD binary inclusion complexes. *Colloids Surf. A Physicochem. Eng. Asp*. **2021**, *611* (2), 125775. DOI: 10.1016/j.colsurfa.2020.125775.
21. Tuszyński, P. K.; Szlęk, J.; Polak, S.; Jachowicz, R.; Mendyk, A. In vitro-in vivo correlation (IVIVC): from current achievements

towards the future. *Dissolut. Technol*. **2018**, *25* (3), 20–27. DOI: 10.14227/DT250318P20.

22. Shleghm, M. R.; Mircioiu, C.; Voicu, V. A.; Mircioiu, I.; Anuta, V. Estimation of the in vivo release of amiodarone from the pharmacokinetics of its active metabolite and correlation with its in vitro release. *Front. Pharmacol*. **2021**, *11*, 621667. DOI: 10.3389/fphar.2020.621667.