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Transfer of Analytical Method's Pharmacopoeias for Assay of Pharmacokinetic In-vitro and Impurities Profile Based on Different Concepts

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Abstract

This paper was carried out to give requirements , criteria and protocol for a transfer (re-validation/partial validation) of pharmacopeia analytical methods based on European Pharmacopeia (EP) and United State Pharmacopeia (USP) requirements. The different steps in a transfer of analytical methods for quantification of active ingredient (assay), impurities profile of drugs sub-stance and finish products and assay of drug dissolution assay (pharmacokinetic in-vitro) were explored, discussed and illustrated with a examples. The recommendations and requirements for this purpose were described in some international guidelines that due to the lack details of protocol. The authors showed both protocols with aid protocol of International Conference harmonization (ICH) and simple statistical process for management of data obtained of transfer of method's pharmacopeia. These analytical methods need verification of system suitability (revalidation or partial validation) , the method's pharmacopeia (reference method) for quality control of drug substance isn't formal partial validation required based on EP concept, while these methods for quality control of drug product need revalidation based on EP and USP requirement. In conclusion the transfer and application of method's pharmacopeias by pharmaceutical manufactures require system suitability test and partial revalidation to assure the report of routine analysis results and reports.

Keywords: *Transfer , re validation , Method's Pharmacopeias .*

1. Introduction

The Pharmacopeias have an essential role in drugs quality control and economical properties that reduce the time for customers [1-3]. Analytical methods pharmacopeia's such as United State Pharmacopeia (USP), European Pharmacopeia (EP) , Japanese Pharmacopeia (JP) and British Pharmacopeia (BP) standards are validated. The suitability of analytical method's pharmacopeia is a very important and contributes in analysis of drugs (e.g. raw materials (drug substance's synthetic way),impurity profiles from different way of synthesis, formulation composition, interference with excipients, manufacture methods for the drug product , matrix effect, chromatographic methods suitability and column appropriateness. Therefore the pharmaceutical international organizations such as EP and USA require users the analytical methods to demonstrate the user's competence to successfully run of analytical methods based on re-validation (called partial validation) [4], where the re-validation can range from as little as one intra-assay accuracy and precision determination to a nearly full validation [5]. The re-validation or partial validation for analytical method's pharmacopeia is the assessment of whether the procedure can be used for its intended purpose, under the actual conditions of use for a specified drug substance and/or drug product matrix. This short communication paper aimed to describe clearly the steps which must be taken into consideration for the re-validation (called transfer of analytical method's pharmacopeia or partial validation in Table 1).

On the other meaning, the paper presented the re-validation of analytical method's compendial for assay of active ingredients, dissolution assay (assay of drug release) and assay of impurities profiles using the EP and USP approach [6,4].

Table 1 : Summary of transfer of analytical method (partial validation) based on EP

Raw materials		**Medicinal product**		
Assay	Impurities profile	Assay	Dissolution test	Impurities profile
System suitability test and no formal validation required	System suitability test and no formal validation required	Specificity Accuracy repeatability Linearity	Specificity Accuracy repeatability Linearity	Specificity Reporting threshold of limit quantification

Note: The analytical method identification isn't formal validation required.

2. Materials and Methods

2.1. Reagents and standard

Standard (reference) of diclofenac sodium (98.2%) , related impurity A standard of diclofenac sodium (100 %) were `obtained from Novartis in Morocco. Mobile phase solvents namely methanol for chromatographic condition (Sigma - Aldrich of Germany). Hydrochloric acid and phosphoric acid (Merck KGaA of Germany). Sodium phosphate tribasic , monobasic sodium phosphate (Riedel–de Haeri of Germany).

2.2. Apparatus

The chromatographic instrumental conditions included the Waters 2695 pump, auto sampler - Waters 2998 photodiode-array detector (PDA). The data acquisition was managed by the Empower Software data registration TM. Hanson SR8-Plus™ of USA . In addition , the Ultraviolet – Visible spectrometer (Perkin of USA), pH meter of Schott of Germany.

2.3. Re-validation / partial validation of method's

Two protocols were used in re-validation of compendial analytical method's with the aid of the EDQM-based EP document (validation of analytical procedure) [4] and USP chapter 1226 (verification of compendial procedure) (Its monograph) [6] .

2.3.1. Chromatographic conditions

The high performance liquid chromatography (HPLC) conditions included the following: 1) 254-nm UV-detector , 2) 4.6-mm×25-cm of column size (packing L7), 3) the mobile phase consisted of a mixture of aqueous phosphate buffer (mix equal volume 0.01 M phosphoric acid and 0.1 M monobasic sodium phosphate at pH 2.5 ± 0.2 and methanol was 30:70 v/v) , the mobile phase was filtered with 0.45-µm Millipore filter and degassed by vacuum. 4) the flow rate was 1.0 ml/minute with injection about 10µl of the standard preparation and the assays preparation into the chromatography, 5) the chromatograms of assay and impurities were recorded, and measure the response of major peaks [6].

2.3.2. Assaying of API in dosage form

Based on the procedure of EDQM, the specificity against the excipients present in the pharmaceutical commercial formulation was re-validated by standard injection and drugs formulations. Also two samples were prepared into an independent way: solutions of active ingredients and solutions of active ingredient in matrix. The samples were prepared in three concentrations , three assays and two replicates in each assay (dissolve weighed DS standard in diluents (70 volume of methanol and 30 volume of water) that included 50 mg/100 ml, 75 mg/100 ml and 100 mg/100 ml with matrix to verify the linearity and accuracy. Also solutions of active ingredients in matrix were prepared as 75 mg of DS in 100 ml in diluents with two assays with three replicates for each assay to verify the precision (repeatability). The samples were shaken mechanically about 10 minutes and sonicated 10 minutes , then a the solution filtered by 0.45-µm filter paper. In addition, the system suitability was tested [7-9]. On the other hand, the repeatability was carried out only for five injections in USP concept (Table 2 and 2).

Table 2 : Linearity of partial validation of HPLC method to assay the API in tablet based on EDQM concept

Assay No	X Introduced (µg/ml)	Y (Area)	X obtained (µg/ml)	Recovery
1 – 1	505	4879161	501.89133	99.38
1 – 2	499	4871364	501.130053	100.42
2 – 1	751	7473658	755.210701	100.56
2 – 2	760	7497183	757.507616	99.672
3 – 1	1001	9999106	1001.78832	100,07
3 – 2	1001	9974288	999.365163	99.83

% RSD of Y was used in the repeatability of USP condition while % RSD of recoveries in EDDM condition

Table 3 : Partial validation data (Accuracy and precision) of HPLC method for assay of API in tablet form based on EDQM concept

Assay No	X Introduced (µg/ml)	Y (Area) *	X obtained (µg/ml)	Recovery
2 – 1	500	4794980	493.672134	98.73
2 – 2	500	4796218	493.793009	98.75
2 – 3	501	4809937	495.132494	98.82
2 – 1	500	4808803	495.021773	99.00
2 – 2	505	4879161	501.89133	99.38
2 – 2	499	4871364	501.130053	100.42

2.3.3. Impurities profile

A solution was prepared with a known concentration of about 0.1 mg/ml of USP - DS related compound in methanol. The HPLC was carried out to determine the related compound A of DS products. In the EDQM, the specificity, system suitability and reporting threshold of limit quantification were determined as re-validation (partial validation) [4], while in the USP concept only the specificity, system suitability were tested [6].

2.3.3. Assaying of drug dissolution testing

In the EDQM procedure, the specificity was verified against the excipients present in the pharmaceutical commercial formulation by scanning the standard and tablet formulations at 1.0 mg/ml. Also the standards samples were prepared in three known concentrations with three assays as the following : dissolve an accurately weighed standard of DS in diluents (dissolution medium), namely 0.01 mg / ml , 0.02 mg/ml and 0.03 mg/ml with matrix (excipient) to verify the linearity. Also the standards were prepared as 0.02 mg/ml of DS in 100 ml in diluents with two assays with three injections for every assay to verify the accuracy and the precision (repeatability) of the analytical method . The samples were shaken mechanically about 10 minutes and sonicated 10 minutes and then a portion of this solution filtered through a 0.45 -µm filter paper. While in the USP , the re-validation (partial validation) of drug release (dissolution testing) is not clear but the specificity (not interference with excipient) and the repeatability (six assays) were carried out (Table 4) .

Table 4 : The linearity of partial validation of spectrophotometric method to assay drug dissolution testing using EDQM concept

Assay No	X Introduced (mg/ml)	Y (Absorbance)	X obtained (mg/ml)	Recovery %
2 – 1	0.01	0.423	0.0102	102.32
2 – 2	0.01	0.415	0.0100	100.30
2 – 3	0.02	0.799	0.0197	98.690
2 – 1	0.02	0.799	0.0197	98.690
2 – 2	0.03	1.21	0.0301	100.43
2 – 2	0.03	1.21	0.0301	100.43

Table 5: The accuracy and precision of partial validation of spectrophotometric method to assay drug dissolution testing based on EDQM concept

Assay No	X Introduced (mg/ml)	Y (Absorbance)	X Obtained (mg/ml)	Recovery %
2 – 1	0.02	0.799	0.0197	98.69
2 – 2	0.0199	0.78	0.0192	96.77
2 – 3	0.02	0.799	0.0197	98.69
2 – 1	0.02	0.799	0.0197	98.69
2 – 2	0.02	0.789	0.0194	97.43
2 – 2	0.02	0.799	0.0197	98.69

The repeatability with the USP concept was expressed by % RSD of response . While in EDDM – based EP concept was expressed by % RSD of recoveries .

2.4. Application

2.4.1 Assay of active ingredients

An accurately weighed portion of powder was transferred equivalent to about 50 mg of DS, to a 100-ml volumetric flask, add about 100 ml of diluents, shaken by mechanical means for 10 minutes and sonicated for about 5 minutes and then a portion of this solution filtered through a 0.45-µm filter paper.

2.4.2. Dissolution testing

The dissolution rate of DS tablet (50 mg) was determined at 37° C in 900 ml of the dissolution medium using a USP six-station dissolution apparatus (Hanson Research of USA) with a paddle rotating at 50 rpm. The dissolution medium was the phosphate buffer (pH 6.8) composed of sodium phosphate tribasic (76 g in litter) and hydrochloric acid (0.1 N). Consecutively, 250 ml and 750 ml were mixed with the pH 6.8 ± 0.05. The drug concentrations in the dissolution samples were analyzed using a UV spectrophotometer at a wavelength of maximum absorbance (276 nm) , the dissolution medium was used as suitable dilution [6,7] .

2.5. Statistical analysis based on different concepts

In both concepts, the data obtained of system suitability of compendial method were analyzed and computed by Empower software of Water's HPLC.

2.5.1. EDQM – based EP approach

A linear regression was fitted. The accuracy was expressed by percentage (%) of recovery (R) at three concentration levels. In addition, the precision (repeatability) was expressed by percentage (%) of relative standard deviation (RSD) at one concentration levels (100%) of the validation standards [4].

2.5.2. USP approach

The precision namely repeatability was expressed in terms of % RSD for two assays with three determinations at target concentration levels (100%) of the validation standards [6].

2.5.3. Re-validation data analysis

The excel software was used to compute the re-validation results of the spectrophotometric method as well as to obtain the linearity, precision and accuracy (expressed by recovery) based on the following equations (1 – 3) [8-12] . .

$$y = ax + b \qquad \text{Eq. (1)}$$

where b is the intercept of the straight line , a is the slope of the line , y is the response (absorbance) and x is the introduced concentration of calcium .

$$\text{RSD (\%)} = \frac{a}{x} \text{x100} \qquad \text{Eq. (2)}$$

$$\text{R (\%)} = \frac{\text{x}^{\wedge}}{\mu^{\wedge}}.100 \qquad \text{Eq. (3)}$$

where, µ^ is the mean of the introduced concentrations and x^ is the estimate of the mean concentration obtained .

3. Results and discussion

3.1. Method's Pharmacopeia Transfer

3.1.1. System suitability

In both approaches, system suitability testing was evaluated by asymmetry factor (AF), retention time (R_T) capacity (k'), theoretical plat and resolution (RS) between DS and impurity A and the results were listed in Tablet 6. The tests were carried out according to the method's pharmacopeia **(Table 6 and Figure 1)**.

Table 6 : System suitability of pharmacopeia analytical method

	Active Ingredient	Imp. A
AF	1.90	1.2
R_T (minutes)	16.02	8.60
k'	1.50	7.50
RS	12.9	

AF: The asymmetry factor is a measure of peak tailing. It is defined as the distance from the center line of the peak to the back slope divided by the distance from the center line of the peak to the front slope, **k'** : It is a measure of the retention of a peak that is independent of column geometry or mobile phase flow rate , **RS** : Resolution is defined as the center-to-center separation between two peaks divided by the average baseline width of those peaks, $\mathbf{R_T}$: is the elapsed time from injection until the highest-concentration part of the peak has eluted from the column [**6**]

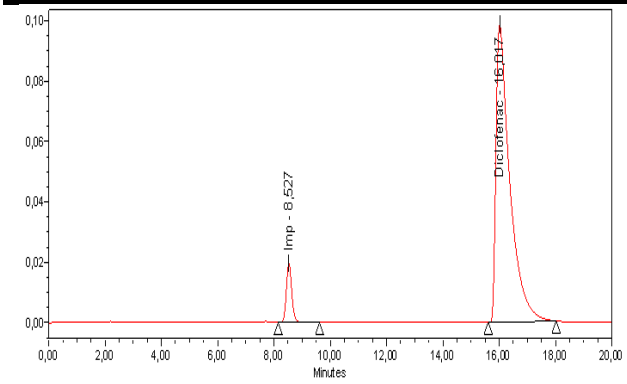


Figure 1. Chromatogram of compendial method to assay of DS and its impurity A

3.1.2. Assay of active ingredients

The RP-HPLC - UV (254 nm) detection was used in order to determine active ingredient of DS content in pharmaceutical commercial tablets. In the EDQM approach, three concentrations standards levels of DS with two repetitions were prepared to evaluate the relationship between the area under the curve and the concentration. The linearity was assessed in a concentration range from 0.05 to 1.0 mg/ml, covering the normal range of concentrations obtained when analyzing content of active ingredient in dosage form with slope 10242, intercept 261210, R^2 equal 0.9998 (**Table 7 and Figure 2**). In addition, only the repeatability of the analytical method was tested by analyzing two assays with three replicate samples of 0.05 mg/ml of DS; the RSD (%) was 0. 65 % . Also the recovery of DS for synthetic mixtures was 99.18. While in based on USP approach, only the repeatability of the method was tested by analyzing six replicate samples ; the RSD was 0. 79 %.

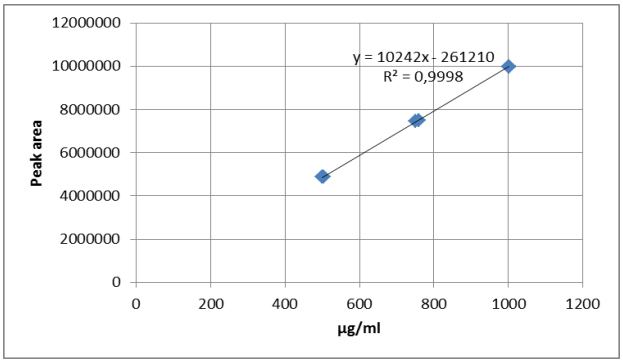


Figure 2. The linearity of HPLC method to assay of DS in tablet

Table 7 : Partial validation parameters of HPLC method to assay the active ingredient using USP and EDQM concept

Characteristics	Assay	
	EDQM	USP
Specificity	+	+
Accuracy (%)	99.18	-
Precision – Repeatability (RSD%)	0.65	0.79
Linearity – R^2	0.9998	-
- Slope	10242	-
- Intercept	2161210	-
• LOQ	-	-

3.1.3. Impurities profile

In EDQM and USP approaches, the system suitability of HPLC was determined to assay of the impurities in pharmaceutical products (related compound A) using USP condition. The major parameters of the system resolution between impurity A and standard was 16.02, retention time, asymmetry factor , capability factor and number of theoretical plat were recorded. In addition, with the EDQM approach , the reporting threshold of limit quantification was found more than 10 about 16 at 5.62 µg/ml (**Table 8 and Figure 3**).

Table 8 : HPLC method re-validation (partial validation) for impurities profile using the USP and EDQM procedure

Characteristics	Impurities Profile	
	EDQM	USP
Specificity	+	+
Accuracy (%)	-	-
Precision – Repeatability (RSD%)	-	-
Linearity – R^2	-	-
- Slope	-	-
- Intercept	-	-
• LOQ	16 at 5.62 µg/ml	-

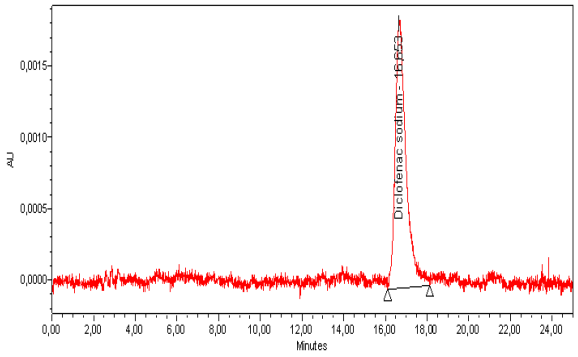


Figure 3. HPLC chromatogram (LOQ , 5.62 µg/ml)

3.1.4. Drug dissolution test/drug release assay

The drug dissolution testing of USP (drug release assay) were assayed to determine the active ingredient release from tablet matrix. In EDQM and USP concepts , the excipients in the pharmaceutical formulation do not interfere with active ingredient (Figure 4). Based on the EDQM (EP), all different concentrations of DS standards were prepared to assess the relationship between the area under the curve and the concentration. The linearity of analytical method was assessed with a concentration range of 0.01 – 0.03 mg, that covering the normal range of concentrations obtained at analyzing % of DS release from tablet with 39.60 of slop , 0.02 of intercept and 0.999 of R^2 (Figure 5). Only the repeatability of spectrophotometric analytical methods was estimated using analyzing six replicate samples of 0.02 mg/ml of DS; the RSD was 0.90 %. The recovery of this method to assay the dissolution of DS was 98.16 %. While in the USP condition, the precision repeatability (RSD%) only was tested with 1.00 % (Table 9).

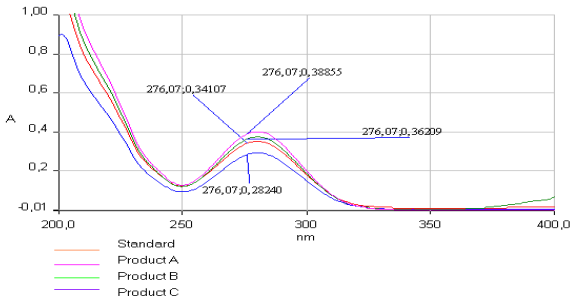


Figure 4 . Specificity of spectrophotometric method for drug dissolution testing of DS tablet dosage form .

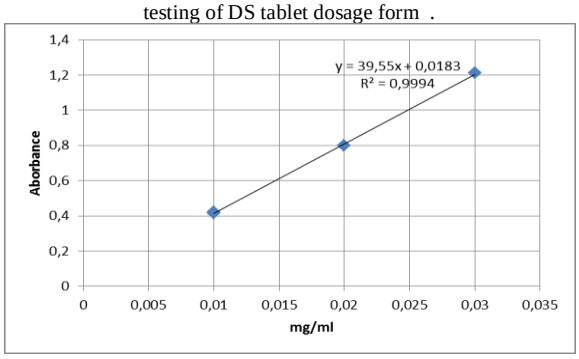


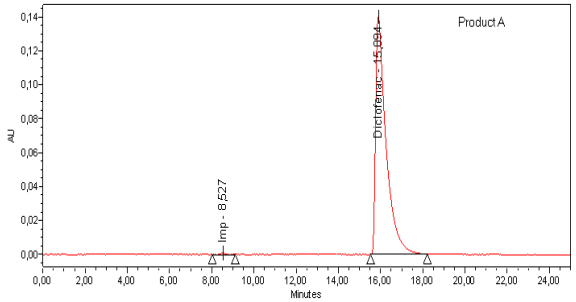
Figure 5 . Linearity of spectrophotometric method to drug release assay (drug dissolution testing) of DS tablet dosage form

Table 9: Partial validation of UV-spectrophotometric technique for assay of drug release (dissolution testing) according to USP and EDQM

Characteristics	Dissolution	
	EDQM	USP
Specificity	+	+
Accuracy (%)	98.20	-
Precision - Repeatability (RSD%)	0.90 %	1.00 %
Linearity - R^2	0.999	-
- Slope	39.60	-
- Intercept	0.02	-
• LOQ	-	-

3.2. Application

The finding of active ingredient content in DS tablets were summarized (See Table 10). On the other mean , the quantity of DS in tablets dosage form within USP normal range (93 % - 107 %) in all products A, B and C (50.43 mg, 50.30 mg and 50.20 mg, respectively). Also, the finding of impurities profile in DS tablet dosage form (active ingredient related compound A) in all products were less than 0.5 % due to USP normal value. On other hand, the results of percentage release of DS (original A, and B, C as generic tablets form marketed in Morocco) at 45 minutes in buffer stage 6.8 using partial validated analytical method have reported in Table 10. The DS products percentages of drug release were confirmed in two products A and B in USP condition (Not less than 75 % (Q)). The failure of active ingredient release matrix of product C was 61.74%. This results due to different in excipients types , physical properties (particles size), or the manufacturing processes used to produce the final pharmaceutical product C.



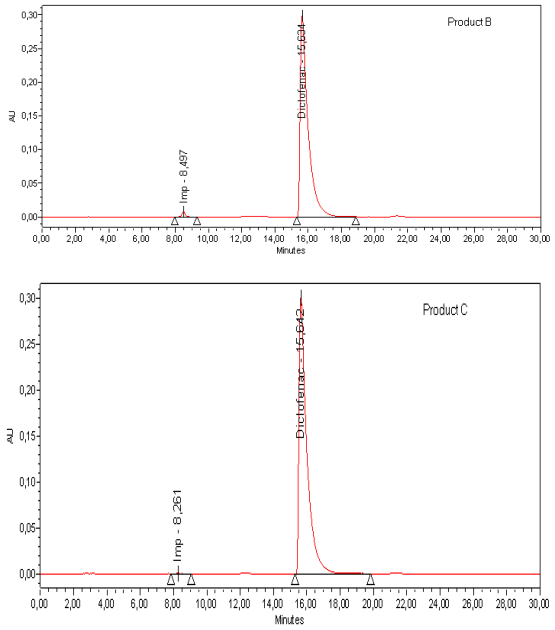


Figure 6. Chromatograms of assay and impurities profile of original product A , generic product and generic product C

Table 10 : Application of analytical methods

	Assay (mg/tablet)	Dissolution (%) at 45 min	Imp. A (%)
Original A	50.40 ± 0.50	84.40 ± 4.20	0.1
Generic B	50.30 ± 0.50	95.30 ± 7.60	0.5
Generic C	50.20 ± 0.2	61.70 ± 6.70	0.1

Briefly, the guideline of EDQM that called validation of analytical procedures gives recommendations and requirements on the extent of the needed re-validation (partial validation) or transfer of analytical methods of pharmacopeia. The EDCM compendial method needs re-validation because this method for specific dosage form is a good basis for the analysis but in several cases there is no indication about the same composition of the product (qualitative and quantitative composition of the excipients). While , in general USP chapter (1226) that called verification of compendial methods. The partial validation or re-validation in USP called verification requirements that should be based on the complexity assessment for both the approaches and the material to which the procedure will be applied. Whereas the complete re-validation of a method's compendial is not required to verify the system suitability , some of the analytical performance characteristics listed in chapter 1225, may be used for the verification process. Only those characteristics that are considered to be appropriate for the verification of the particular method need to be evaluated. The degree and extent of the verification process may depend on the training level and operator experience, the procedure , quality of equipment and instrumentation. Also , steps of specific procedure . In addition, the partial validation criteria available in USP monograph that include the specificity, system suitability and repeatability (RSD %) only . Due to less information about guideline or regulatory requirements, several concepts are possible to select the simple protocol to analytical pharmacopeia transfer and apply in drug quality control laboratories. To choose the statistical data treatment namely the assay of active ingredient , impurities profiles and dissolution test [8-12]. Several terms and statistical approaches for the partial validation of analytical methods have been described and designed. The USP described the partial validation of analytical procedures (verification). This approach gives enough guarantees for the future results that will be generated by this method during routine use will be close enough to the true value [13-15].

5. Conclusion

The study concluded that the analytical procedures of compendia guideline were validated but according to the USP and EP approach give recommendations and requirements on the extent of analytical method's pharmacopeia (partial validation / verification) needed for analytical method .

Data Availability

No data were used to support this study**.**

Conflicts of Interest

The authors declare that they have no conflicts of interest**.**

Acknowledgments

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Ethical Approval

The study were reviewed and approved by Ethics Committee of Laboratory of Pharmacology and Toxicology, Faculty of Medicine and Pharmacy – University Mohammed V - Soussi , Rabat, Morocco **.**

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References :

[1] Artiges A.,. The role of pharmacopoeias in international harmonization. Journal of Pharmaceutical and Biomedical Analysis . (**2001**) 24, 769–772 .

[2] Calam D.H ., Design and international harmonization of pharmacopoeia1 standards . Journal of Pharmaceutical and Biomedical Analysis . (**1995**) 14 , l-5 .

[3] International Conference on Harmonization of Technical Requirement for the Registration of Pharmaceuticals for Human Use Topic Q4 (R1) : Pharmacopoeias , (**2007**).

[4] European Directorate for the Quality of Medicine and Health Care. Validation of analytical procedure (**2005**)

[5] Food and Drugs Administration . Center for Drug Evaluation and Research (CDER) , Validation of Bioanalytical Method Methods (**2001**) .

[6] Unites State Pharmacopeia (USP). Unites States Pharmacopoeia Convention . 33 (**2010**).

[7] Amood Al Kamarany, M., Karbane , M. E., Alanazi , F., Cherrah , Y., & Bouklouze , A. (2023). Effect of Biopharmaceutical Classification System on Pharmacokinetic and Mechanism of Drug Release In vitro . Abhath Journal of Basic and Applied Sciences, 2(2), 49–53. https://doi.org/10.59846/ajbas.v2i2.524

[8] International Conference on Harmonization of Technical Requirement for the Registration of Pharmaceuticals for Human Use Topic Q2 (R1) : Validation of analytical procedures : Text and Methodology , Geneva , (**2005**).

[9] El Karbane, M.; Azougagh, M.; Amood, A.L.K.M.; Bouchafra, H.; Cherrah, Y.; Bouklouze, A. Development and validation of a reversed-phase HPLC method for simultaneous analysis of butylhydroxyanisol, simvastatin and its

impurities in tablet dosage forms. Ann. Pharm. Fr. 2014, 72, 244–255.

[10] Amood Al Kamarany, M., & Abary , M. Y. S. (2023). In vitro – In vivo Correlation : Dissolution Profile and Pharmacokinetic of Calcium Carbonate Tablets Marketed in Yemen. *Abhath Journal of Basic and Applied Sciences*, 2(2), 54–60. https://doi.org/10.59846/ajbas.v2i2.525

[11] Al-Kamarany, M. A., Karbane, M. E., Ridouan, K., Alanazi, F. K., Hubert, P., Cherrah, Y., & Bouklouze, A. (2012). Transfer of drug dissolution testing by statistical approaches: Case study. *Saudi Pharmaceutical Journal*, 20(1), 93-101.

[12] Al-Kamarany MA, Alwosaby AS, El Karbane M, Karrouchi K. (2013) SFSTP guideline 1992- based validation of direct potentiometric method for quantification of sodium bicarbonate in baby's bliss gripe water available in Yemen. *Int J Pharm Pharm Sci* 5: 411-413.

[13] Elkarbane, M., Al-Kamarany, M. A., Bouchafra, H., Azougagh, M., Cherrah, Y., Hubert, P., & Bouklouze, A. (2015). Total error-based validation including the experimental design-based robustness evaluation of a stability-indicating method for the simultaneous quantification of hydrochlorothiazide and valsartan in tablet formulations. *Acta Chromatographica*, *27*(2), 195-215.

[14] Karbane, M. E. L., Ramli, Y., Al-Kamarany, M. A., Bouchfra, H., Radi, M., Karrouchi, K., ... & Azougagh, M. (2014). Development and validation of HPLC dissolution assay of simvastatin tablets under normal and accelerated conditions. *Journal of Chemical and Pharmaceutical Research*, 6(6), 886-893.

[15] Al-Kamarany, M. A., El Karbane, M., Belomaria, M., Azougagh, M., Hubert, P. H., Alanazi, F., & Bouklouze, A. (2012). Development and validation of LC method for the diclofenac sodium release determination using accuracy profile concept. *Asian Journal of Chemistry*, 24(10), 4393.