Arabian Journal of Chemistry (2017) **10**, S3245–S3254

King Saud University

Arabian Journal of Chemistry

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ORIGINAL ARTICLE

A validated stability-indicating UPLC method for determination of diclofenac sodium in its pure form and matrix formulations

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Received 26 February 2013; accepted 22 December 2013
Available online 31 December 2013

KEYWORDS

Ultra performance liquid chromatography;
Diclofenac sodium;
Forced degradation;
Diclofenac related compound A;
Stability-indicating assay

Abstract The aim of this work is to develop a validated stability indicating reverse phase ultra performance liquid chromatography (UPLC) method for the rapid and accurate determination of diclofenac sodium in its pure form and in matrix formulations. This UPLC method is composed of isocratic mobile phase, 0.05 M ammonium acetate buffer (pH = 2.5) and acetonitrile (50:50), with flow rate 0.5 ml/min, column BEH C18 (2.1 × 50 mm, 1.7 μm). The method is rapid (1.2 min run), selective with well resoluted diclofenac peak with retention time 0.94 min and sensitive (LOD = 2 ppm and LLOQ = 6 ppm) with UV detection at 254 nm. The drug was subjected to acidic, alkaline media, boiling and oxidizing agent to apply stress conditions. The developed method was able to separate degradation product generated under forced degradation studies. The developed method was validated as per the FDA guidelines for specificity, linearity, accuracy, precision, LOD, LLOQ and found to be satisfactory. The study suggests that the developed UPLC method can be used for the assessment of drug purity and stability. It can be also used to monitor the drug content and release from different formulations without any interference of excipients and/or degradation products.

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Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

1. Introduction

Diclofenac sodium (DS) (Fig. 1) is sodium-[(2,6-dicholorophenyl)amino] – phenyl acetate. It is a sodium salt of an arylacetic acid derivative (Tripathi, 1995; United States Pharmacopoeial Convention, 2000). It inhibits prostaglandins synthesis by interfering with the action of prostaglandin synthetase (Cyclooxygenase) (Tripathi, 1995). It possesses analgesic, anti-inflammatory and antipyretic activity. It is widely used

http://dx.doi.org/10.1016/j.arabjc.2013.12.022
1878-5352

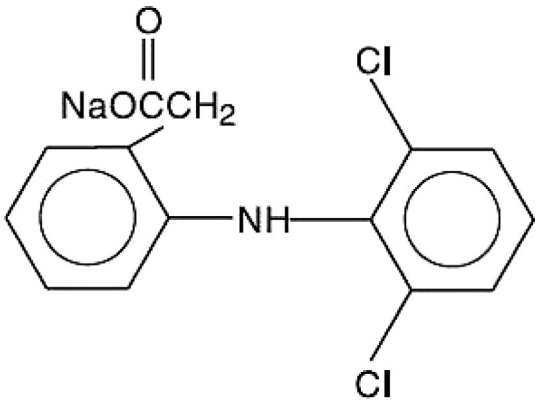

Figure 1 Chemical structure of diclofenac sodium.

in various clinical disorders including rheumatoid arthritis and osteoarthritis, soft tissue disorders, renal colic, acute gout, dysmenorrhea and migraine (Sweetman, 2009).

Ultraviolet spectrophotometric methods are often employed in the analysis of pharmaceutical preparations containing DS (Agrawal and Shivramchandra, 1991; Fabre et al., 1993; Kamath and Shivram, 1993) with high performance liquid chromatography (HPLC) being used in its evaluation in tablets (Beaulieu et al., 1990; Kubala et al., 1993). Many HPLC methods with UV detection were adopted in the literature for the determination of DS (El-Kommos et al., 2012; Hafsa et al., 2011; Kole et al., 2011; Nasir et al., 2011; Panda et al., 2012; Raman and Patil, 2001; Sultana et al., 2011). HPLC provides more accurate determination of the drug than using spectrophotometric methods especially in the presence of interfering matrices resulting from different excipients in different formulations. However, most of the available HPLC methods may not be the analytical methods of choice when it is required to increase the throughput and reduce analysis cost as compared to UPLC methods. For the routine quality control work, hundreds of samples have to be analyzed on daily basis consuming a huge amount of time and resources "solvents and equipment". UPLC methods being simpler, faster and more economic methods of detection and quantification than HPLC, they represent an important alternative. UPLC is relatively new technique that ensures a decrease in run time and solvent consumption. This system can simply withstand high system back-pressures during chromatographic analysis without negatively influencing the analytical column (Novakova et al., 2006). Sample injection is characterized by fast injection cycles, low injection volumes, and negligible carryover.

A literature survey reveals that there is no method reported so far for UPLC determination of DS using simple UV detection. In the present work, a stability indicating, simple, economic, rapid, precise and sensitive reverse phase UPLC method was proposed and validated for determination of DS in its bulk form, slow release HPMC K15M matrix tablets and Eudragit L 30D-55 enteric coated pellets. This method would facilitate effective and economic management of resources especially for quality control (QC) aspects.

2. Experimental

2.1. Materials and supplies

An authentic diclofenac related compound **A** USP standard was purchased from USP. An authentic USP reference standard DS was generously provided by the Saudi FDA. DS (assay 99.85% w/w) was generously provided by TABUK Pharmaceuticals, Tabuk, Saudi Arabia. Sodium chloride and potassium dihydrogen phosphate were purchased from Merck KGaA, Darmstadt, Germany. Potassium chloride was obtained from Fluka Chemicals AGCH, Switzerland. Disodium phosphate dodecahydrate and acetonitrile (HPLC grade) were obtained from BDH, UK. Deionized water was obtained in-house through ELGA® purification system, Vivendi Water Systems Ltd., UK and was further filtered through a 0.2 μm membrane PTFE Disposable syringe filter from Macherey–Nagel GmbH, Germany.

2.2. Instrumentation and chromatographic conditions

Chromatography was performed on Acquity® (UPLC) system equipped with Binary Solvent Manger, Acquity® automatic sample manager, column heater, Acquity® photodiode array (PDA) eλ detector and Acquity® UPLC BEH C18 column (2.1 × 50 mm,1.7 μm) obtained from Waters (Waters Inc., Bedford, MA, USA).

Separation employed reverse-phase isocratic elution using a mobile phase consisting of 0.05 M acetate buffer (pH, 2.5) and acetonitrile (50:50, v/v) run at flow rate of 0.5 ml/min and injection volume 1 μl. PDA Detector was set to acquire 3D data from 210 to 280 nm while the 2D channel was recording at 254 nm. The column temperature was kept at 50 °C while sample temperature was kept at 10 °C.

2.3. System suitability

System suitability was checked for the conformance of suitability and reproducibility of chromatographic system for analysis. System suitability was determined before sample analysis from five replicate injections of the standard solution containing 1.2 mg/mL of diclofenac sodium. The acceptance criteria were less than 2% relative standard deviation (RSD) for peak areas, USP tailing factor (T) less than 2.0, USP plate count (N) more than 5000, capacity factor (K) more than 2 and resolution (R_s) more than 1.5 for DS peaks from standard solution. All critical parameters tested met the acceptance criteria (Table 1).

Table 1 System suitability results.

Parameters	Acceptance criteria	Actual value	Indication
Area (% RSD, $n = 5$)	⩽2.0	0.632	Good injection repeatability
Plate count (N)	> 5000	8864	Good column efficiency
Tailing factor (T)	⩽2.0	1.0625	Good peak symmetry
Capacity factor (K')	> 2	3.18	The peak is well resolved with respect to the void volume
Resolution (R_s)	> 1.5	2	Good separation of the drug from the degradation product

2.4. Standard solutions

DS stock solution was prepared in 0.2 M phosphate buffer; pH 6.8 to produce final concentration of 2500 μg/ml. Standard solutions were then serially diluted to eleven working standard solutions covering the range (1–2500 μg/ml). This wide concentration range was designed to compass different drug formulations doses. Calibration curves were obtained by plotting peak area against standard drug concentration and regression equations were computed.

2.5. Drug extraction from formulations

DS was extracted from the matrix formulation (HPMC matrix tablets and enteric coated non pareil seeds (40–60 mesh size)) via dissolution in mixture of water and methanol (30:70) and sonicating for 15 min, and then centrifuged at 5000 rpm for 5 min. The clear supernatant is further filtered through a 0.45 μm filter then injected (1 μl) into the UPLC.

2.6. Method validation

Several performance parameters were designed to be tested via validation experiments including specificity, linearity, limit of detection (LOD), lower limit of quantitation (LLOQ), accuracy and precision according to FDA guidelines.

2.6.1. Specificity

It is the ability of an analytical method to differentiate the analyte in the presence of other components in the sample and quantify it. Specificity was assessed to test the effect of matrix for interference at the same retention time as well as to ensure the validity of the method to be further utilized as a stability-indicating assay.

To evaluate the specificity of the method, drug free quality control zero samples were carried out through the assay procedure and the retention times of the matrix formulation components were compared with that of DS analyte. The specificity of the method toward the intact drug was also studied by determination of the resolution (R) between the drug peak and the nearest degradation product (Dongre et al., 2008).

2.6.2. Calibration and linearity

Suitable volumes of DS working standard solution were conducted to prepare non zero standard drug concentrations ranged from (1 to 250 μg/ml) and eight QC concentrations covering the same range including zero concentration (QC_{Zero}). QC samples were prepared by spiking the matrix formulation with known amount of DS, then diluting the mixture to appropriate volume. A calibration curve was constructed from samples covering the total range, including LLOQ. Calibration samples were injected from low to high concentration into 6 replicates for three consecutive days for validation. Linear regression equation and correlation coefficient (R^2) were employed to statistically evaluate the linearity of the results (Al-Hadiya et al., 2010; Ali et al., 2006).

2.6.3. Recovery studies

The recovery of a QC sample is the percent resulted from comparing the response obtained from known concentration of the analyte added to, and extracted from the matrix, and the response obtained for the true concentration of the pure standard. The absolute recovery of DS was evaluated by comparing drug peak area of the QC samples to aqueous standard solution that has been injected directly into UPLC system. The assay of absolute recovery at each concentration was computed using the following equation:

$$\text{Absolute recovery} = \frac{\text{peak area of extract}}{\text{Mean peak area of direct injection}} \times 100 \quad (1)$$

While the relative recovery was calculated by the following equation:

$$\text{Relative recovery} = \frac{\text{Concentration of extract}}{\text{Theoritical concentration}} \times 100 \quad (2)$$

2.6.4. Accuracy and precision

The accuracy of an analytical method describes the closeness of mean test results obtained by the method to the true value (concentration) of the analyte. The precision of an analytical method describes the closeness of individual measures of an analyte when the procedure is applied repeatedly to multiple aliquots of a single homogeneous volume of matrix.

The intra-day accuracy and precision were determined by six replicate analysis of samples at different concentrations within the same day including LLOQ while, the inter-day accuracy and precision were determined by comparing QC analysis performed on three different days repeatedly as sextets of each QC level. The precision was calculated as the relative standard deviation (RSD) and the accuracy of the method was expressed in terms of % drug recovered (Al-Hadiya et al., 2010).

2.7. Limit of detection (LOD) and lower limit of quantification (LLOQ)

The limit of detection and lower limit of quantification were calculated based on the determination of the slope of the calibration curve and the standard deviation (SD) of responses using the following equations:

$$\text{LOD} = 3\frac{\text{SD}}{\text{Slope}} \quad (3)$$

$$\text{LLOQ} = 10\frac{\text{SD}}{\text{Slope}} \quad (4)$$

2.8. Stability-indicating study

2.8.1. Separation of diclofenac related compound A and standard diclofenac sodium

To check for the capability of the method to separate the drug from its related compounds, standard mixture solutions of diclofenac related compound **A** USP standard and diclofenac sodium standard were prepared together in ratios of 1:10 (80 μg/ml:800 μg/ml) and 1:100 (8 μg/ml:800 μg/ml) where diclofenac related compound **A** was of the smaller concentration and a volume of 1 μl was injected into the UPLC system.

2.8.2. Forced degradation studies

The ICH guideline entitled stability testing of drug substances and products (ICH Guideline, 2005) requires the stress testing

to be carried out to elucidate the inherent stability characteristics of the active substance, and provide a rapid identification of differences that might result from changes in the manufacturing processes or source sample. An ideal stability-indicating method is one that quantifies the standard drug alone and also resolves its degradation products. DS (USP reference standard, LOT H0B150) was subjected to different stress conditions including boiling, acid, base hydrolysis and oxidation.

2.8.2.1. Boiling. 10 ml of DS (USP reference standard) solution (2.5 mg/ml) was transferred to a small rounded flask. The solution was subjected to reflux for 2 h in a boiling water bath. The solution was cooled to room temperature (25 ± 5 °C), and a volume of 1 µl was injected into the UPLC system (Kadi et al., 2011).

2.8.2.2. Acid and alkaline hydrolysis. 10 ml of DS (USP reference standard) solution (2.5 mg/ml) was transferred to a small rounded flask. Sufficient quantity of 1 N hydrochloric acid solution was added to reach pH about 1. Similarly, to another 10 ml sufficient amount of 1 N sodium hydroxide was added to reach pH about 12. The prepared solutions were subjected to reflux for 2 h in a boiling water bath. Each degradation sample was cooled to room temperature (25 ± 5 °C), diluted with phosphate buffer pH = 6.8 (1:10). From the resulting solution, 1 µl was injected into the UPLC system.

2.8.2.3. Oxidation. 10 ml of DS (USP reference standard) solution (2.5 mg/ml) was transferred to a small rounded flask. The contents were then mixed with 1 ml of 30% hydrogen peroxide solution, and the reaction mixture was allowed to proceed at room temperature (25 ± 5 °C) for 2 h with intermittent shaking. A volume of 1 µl was injected into the UPLC system.

2.9. Identification of the degradation product using mass spectrometry

The acid treated sample was allowed to run on the UPLC under the formerly described analytical conditions. Once the peaks of the degradation product and the intact drug were resolved from each other, they were collected manually form the eluent to be injected into the mass spectrometer to identify and confirm the nature of the degradation product. Chromatographic injection for each sample was performed on an Agilent 6320 Ion trap mass spectrometer (Agilent technologies, USA) equipped with an electrospray ionization interface (ESI). Direct infusion was conducted using infusion pump. Infusion rate was 5 ul/min. Mobile phase is composed of HPLC grade water and acetonitrile in the ratio 1:1. MS parameters were optimized for each compound. The scan was ultra-scan mode. For screening of mass signals of the different compounds and to search for parent ions for MS/MS experiments, MS2 scans were performed in the mass range of m/z 50–400. The

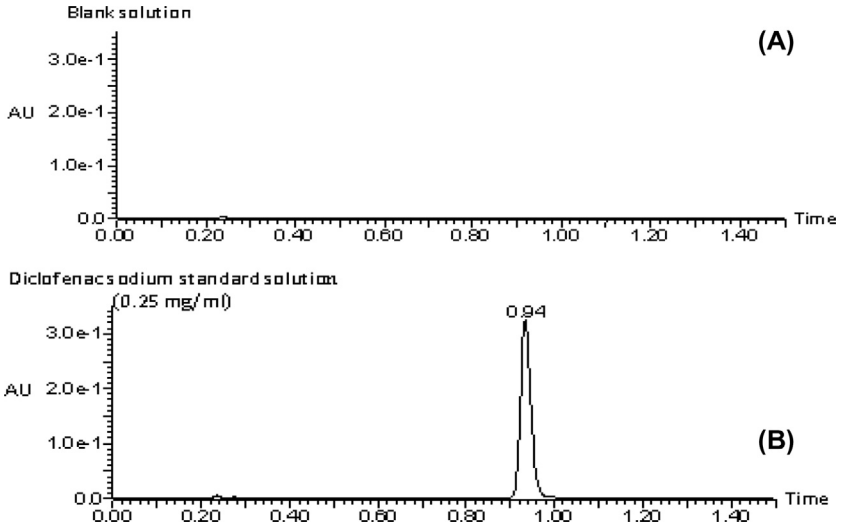

Figure 2 Chromatogram resulting from (A) Blank sample, and (B) Standard DS solution (0.25 mg/ml).

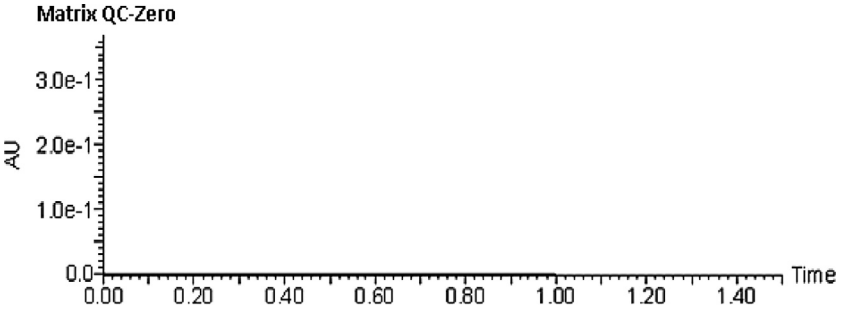

Figure 3 Chromatogram resulting from QC zero.

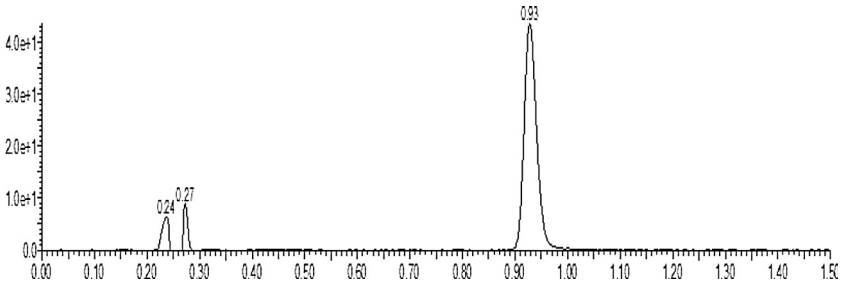

Figure 4 LLOQ chromatogram resulting from matrix formulations spiked with 2 μg/ml diclofenac sodium.

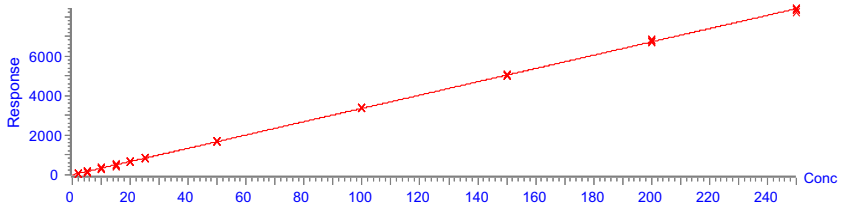

Figure 5 Typical calibration curve of DS standard solutions covering the range (2–250 μg/ml). Response is calculated as peak area against concentration.

ESI was operated in negative mode for the drug and positive mode for the degradant. The source temperature was set to 350 °C nebulizer gas pressure of 55.00 psi; dry gas flow rate of 12.00 L min^{-1}.

2.10. Statistical analysis

The data were analyzed by one way ANOVA followed by the Bonferroni multiple comparison test, using PASW Statistics 18 Software, v. 5.01 (SPSS Software, Inc.,).

3. Results and discussion

The method has been validated for linearity, specificity, recovery, accuracy and precision and found to be within acceptable limits as per FDA guidelines for bioanalytical method validation.

3.1. Separation and selectivity

Specificity was tested in six different randomly selected samples of drug free matrix used for analysis. Fig. 2 shows the representative chromatograms of blank solution (A) and standard DS solution (0.25 mg/ml) (B) compared to each other. Fig. 3 shows the chromatogram of QC_{Zero} sample while Fig. 4 shows the chromatogram of DS at LLOQ level in matrix. The analytes were well separated from matrix samples after subjecting to the same chromatographic conditions at retention time of 0.94 min. The DS peak (R_t = 0.94 min) was of good shape and completely resolved from the nearest background peaks as in Fig. 4.

3.2. Linearity

The peak area of DS was linear with respect to the analyte concentration over the range 1–2500 μg/ml. The mean linear regression equation of calibration curve for analyte was $Y = 33.7097(\pm 0.1786)\,X - 25.8071\,(\pm 2.421)$. The correlation factor (R^2) was 0.9993 ± 0.0011 over this wide concentration range as shown in Fig. 5.

Table 2 Data of back calculated diclofenac sodium concentration of calibration standards.

Nominal concentration (μg/ml)	Mean[a] (μg/ml)	Precision (RSD %)	Accuracy (%)
6	6.12	0.67	102
10	9.78	3.73	97.83
15	15.25	2.86	101.67
20	19.87	2.46	99.33
25	25	2.44	100
50	53.63	0.46	98.79
100	102.07	1.07	102.07
150	152.52	1.18	101.97
200	201.15	2.68	100.58
250	248.28	0.89	99.31

[a] Average of six determinations.

3.3. Limit of detection and lower limit of quantification

The lower limit of detection was estimated to be 2 μg/ml. The LLOQ is the lowest concentration of the standard curve which can be measured with acceptable accuracy and precision for the analyte from the matrix formulation. LLOQ was calculated to be 6 μg/ml, with recovery 103.5% and precision of 0.66%. Table 2 summarizes the back calculation of DS calibration standards.

3.4. Recovery

Relative recovery of the analytes was calculated by comparing the concentration of the extracted drug to the theoretical concentration. Results are summarized in Table 3.

The average recoveries of DS extracted from matrix formulation were between (103.5% and 91.44%) with coefficient of

Table 3 Data of relative recovery of diclofenac sodium from matrix systems.

Sample No.	Nominal concentration (μg/ml)	Mean found concentration[a] (μg/ml)	Recovery (%)	Precision (%)
QC-1	6	6.22	103.5	0.66
QC-2	10	9.4	94	0.67
QC-3	20	20.32	101.58	2.57
QC-4	50	50.45	100.90	0.35
QC-5	100	96.45	96.45	0.59
QC-6	150	137.17	91.44	1.90
QC-7	200	194.08	97.03	0.70
QC-8	250	248.28	99.31	0.89

[a] Average of six determinations.

Table 4 Data of inter-day accuracy and precision of diclofenac sodium QC samples.

Day of analysis	Diclofenac sodium concentration measure in matrix formulations (μg/ml)						
	LLOQ	QC-2	QC-3	QC-4	QC-5	QC-6	QC-7
	6	10	20	50	100	150	200
Day 1	6.2	9.5	20	53.3	96	141.7	213.6
	6.2	9.4	19.7	50.8	95.5	139.5	195.3
	6.2	9.4	19.8	54.6	95.7	139	193.3
	6.2	9.4	19.8	52.0	96.2	137.3	195.0
	6.2	9.3	19.9	50.3	96.3	133.5	194.6
	6.3	9.4	19.8	55.1	96.4	131.6	196.9
Day 2	6.5	9.6	20.9	48.5	96.9	137.5	195.3
	6.5	9.6	20.9	51.0	97.1	136.8	193.9
	6.4	9.5	20.9	50.1	96.2	136.7	192.7
	6.5	9.4	20.5	50.3	97.1	137.6	192.4
	6.5	9.5	20.7	50.4	97.2	137.3	192.7
	6.4	9.5	20.9	50.1	96.8	137.5	193.5
Day 3	6.4	9.8	20	50.3	101.3	149.4	193.3
	6.5	9.6	20.2	50.6	100.9	151.2	204.1
	6.5	9.6	20.4	50.7	101.4	149.5	200.1
	6.5	9.6	20.2	50.5	100.4	149.6	201.0
	6.5	9.7	20.2	50.3	100.7	150	199.9
	6.5	9.6	20.0	50.3	101.3	149.4	203.0
Mean (μg/ml)	6.39	9.52	20.5	50.3	98.9	143.5	196.8
Accuracy (%)	106.5	95.2	102.4	100.5	98.9	95.7	98.4
Precision (RSD %)	2.07	1.33	1.8	1.2	2.2	4.6	2.3

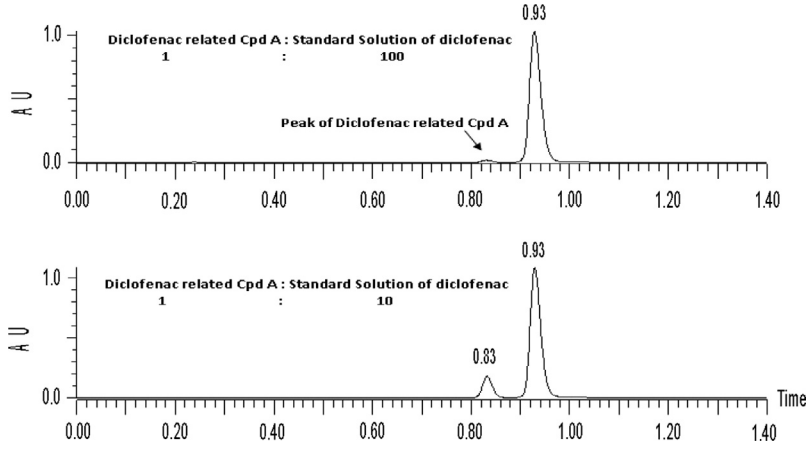

Figure 6 Chromatogram showing peaks of diclofenac related compound (Cpd) A USP standard and standard diclofenac sodium well resoluted from each other in ratios 1:100 and 1:10.

variation between (0.35% and 2.57%). These values were acceptable according to FDA guidelines.

3.5. Precision and accuracy

Inter and intraday accuracy and precision at LLOQ and range containing low, medium and high concentrations of DS in the matrix formulation were within acceptable limits. Intraday and inter-day accuracy range was (97.83–102.07%) and (95.2–106.5%), respectively. Precision (RSD) within-run, intra-batch precision or repeatability was less than 3.73 and 0.46, and found to be less than 1.2 and 4.6 for between-run, inter-batch precision or repeatability. These results are summarized in Tables 2 and 4.

3.6. Stability-indicating study

Fig. 6 shows the two peaks of diclofenac related compound **A** USP standard and that of standard diclofenac sodium well resoluted from each other with the degradation peak appeared at retention time 0.83 min. The peak can be detected at very low concentration compared to the drug concentration even at a ratio 1:100 (Fig. 6). DS (R_t = 0.94 min) is observed to be well resolved from the degradation peak (R_t = 0.83 min). *R* value

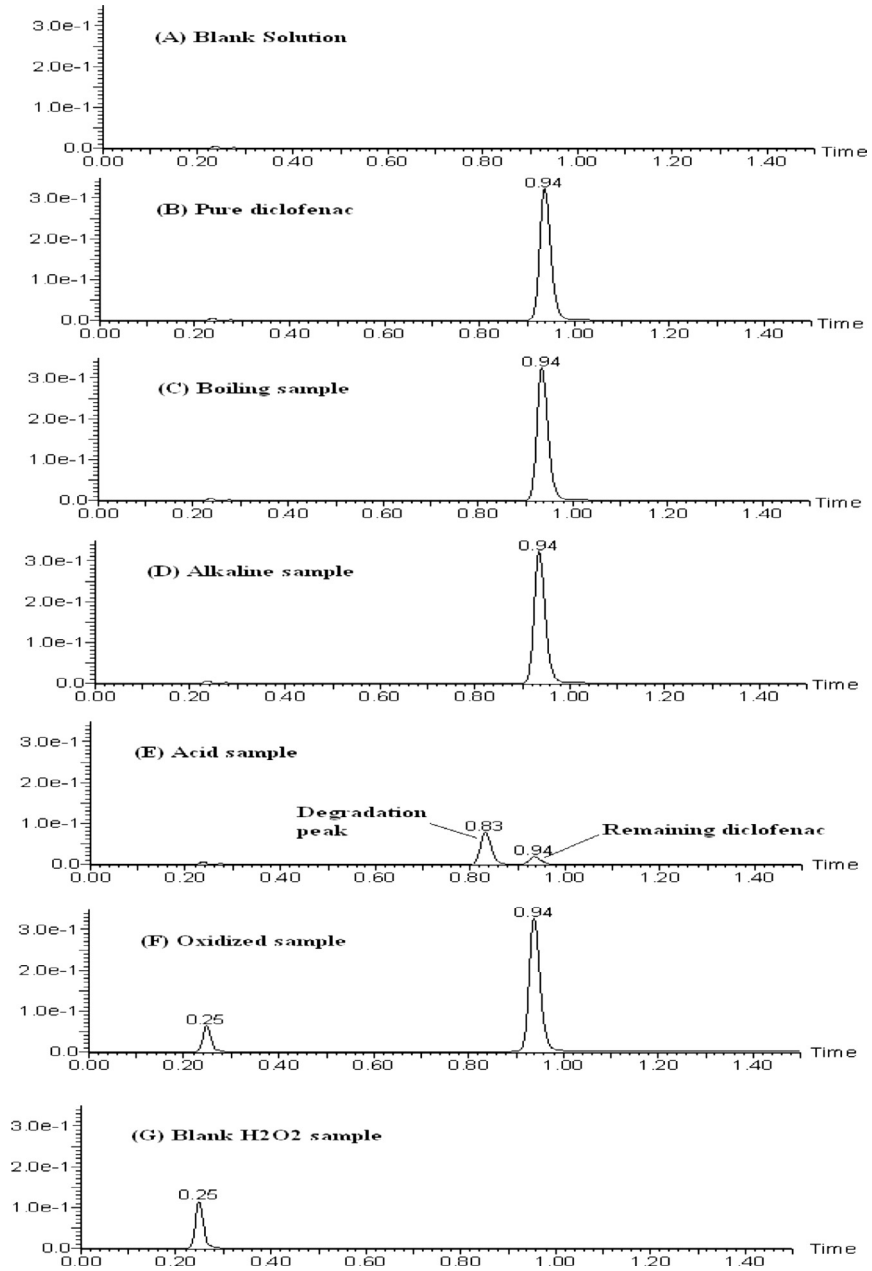

Figure 7 Sample chromatograms of diclofenac sodium and its degradation product. (A) Blank solution, (B) freshly prepared solution of DS (0.25 mg/ml), (C) boiled sample of DS, (D) alkali treated sample of DS, (E) acid treated sample of DS, (F) oxidized sample of DS, (G) Blank solution of H_2O_2.

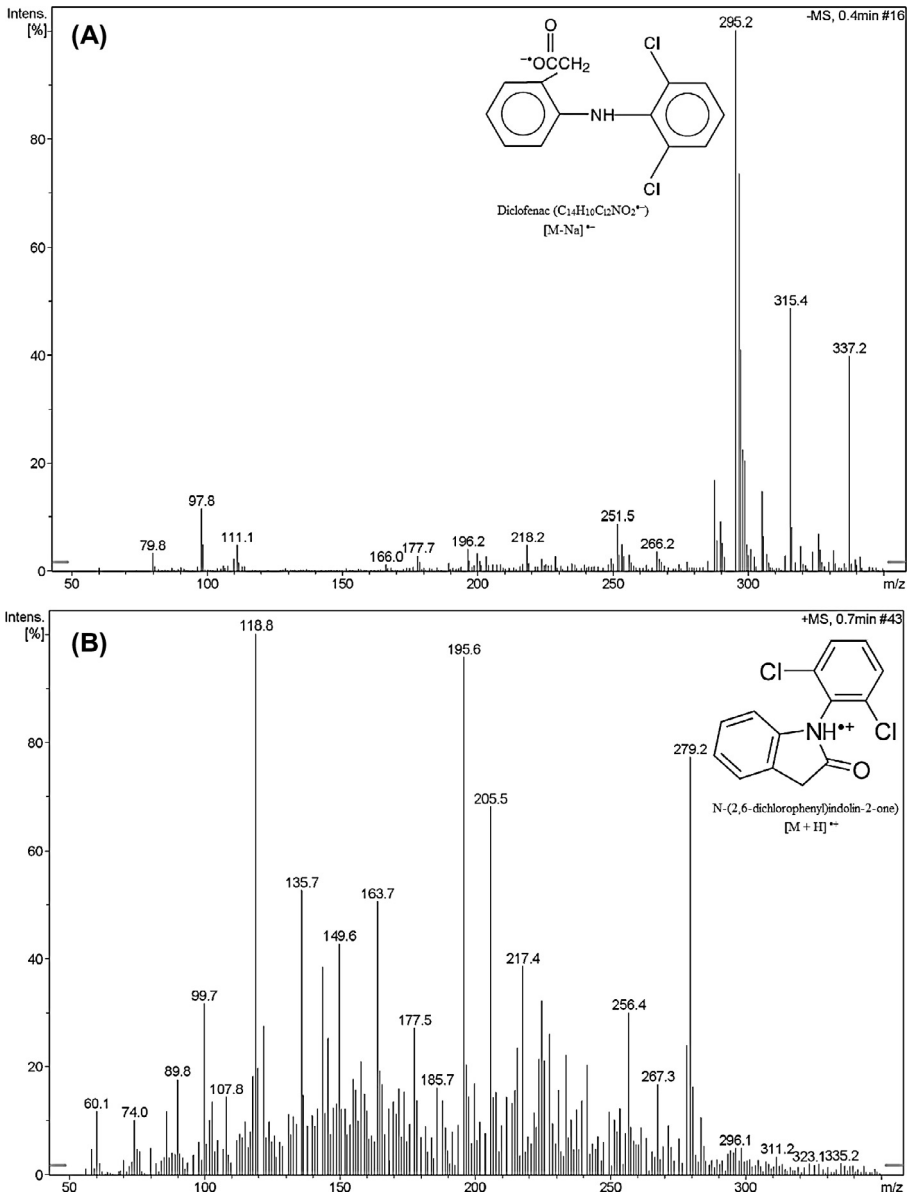

Figure 8 Mass spectrum of (A) diclofenac and (B) the degradation product: diclofenac related compound **A** (N-(2,6-dichlorophenyl)indolin-2-one).

was calculated based on measuring peak width at half-height (USP, 2003)

$$R = \frac{2(t_2 - t_1)}{1.7\left(W_{1,\frac{h}{2}} + W_{2,\frac{h}{2}}\right)} \quad (5)$$

where R denotes the resolution, t_1 and t_2 are the retention times of two peaks and $W_{1,h/2}$ and $W_{2,h/2}$ represent the peak width at half-height of the two peaks. Resolution value between DS and its degradation product was 2 which is considered to be satisfactory.

Forced degradation samples were used also to validate the stability-indicating capacity of the method. The drug seems to be stable in boiling, alkaline and oxidation stress conditions as no degradation products were observed in their chromatograms, which were identical to the chromatogram of DS solution that has not been subjected to any stress conditions (Fig. 7B–D and F). The amounts recovered were 100.2%, 99.75%, and 99.55% for boiling, alkaline and oxidation conditions, respectively. However, DS has degraded to 7.04% of the original concentration after exposure to acidic conditions (Fig. 8E) with peak area 473 compared to 6883 of the intact drug. The degradation peak appeared at the 0.83 min which is the same retention time of diclofenac related compound **A** (N-(2,6-dichlorophenyl)indolin-2-one). The peak area of the degradant was 2038 which represents 29.6% of the area of the intact drug. The relative retention time (RRT) of the degradant peak was calculated (Eq. 6) to be 0.71 indicating that its peak eluted before that of the intact drug.

$$\mathrm{RRT} = \frac{(t_2 - t_0)}{(t_2 - t_0)} \quad (6)$$

where t_0 is the void time, t_2 is the retention time of the degradant and t_1 is the retention time of the intact drug.

3.7. Identification of the degradation product using mass spectrometry

ESI was operated in negative and positive ion modes for the analysis to provide optimum sensitivity and selectivity. The negative mass spectrum Fig. 8(A) for the peak eluting at retention time 0.94 min showed a predominant single ion at m/z 295.1 which represents the molecular ion peak for the parent drug $[M-Na]^{\bullet -}$ (Diclofenac ($C_{14}H_{10}C_{l2}NO_2^-$)) at the same retention time. While the positive mass spectrum Fig. 8(B) for the peak eluting at retention time 0.83 min showed a single ion at 279.2 which corresponds to the protonated molecular ion peak $[M+H]^{\bullet +}$ of diclofenac related compound **A** (N-(2,6-dichlorophenyl)indolin-2-one)), M. Wt. = 278.13 at the same retention time. This degradation compound resulted from the condensation of the carboxylic- and the amino-group in the molecule of diclofenac forming a nitrogen containing ring in the molecule (Reddersen and Heberer, 2003). It can be assumed, that the acid treatment of the sample caused a rapid ring closure reaction (Hartmann et al., 2008). These results confirm the ability of the developed UPLC method to separate and detect the degradation product in the presence of the intact drug.

3.8. Comparison of the proposed reverse phase UPLC method with a conventional reverse phase HPLC (Rp-HPLC) method

The current developed method was compared to a previously published Rp-HPLC method that utilized similar mobile phase composition using a suspect $ODSC_{18}$ (25 cm × 4.6 mm id)with (5 μm particle size) and isocratic elution with a mobile phase containing 16% acetonitrile in 0.02 M sodium acetate buffer pH(5.5) at flow rate of 1 m min^{-1}, 20 μl sample loop, temperature 30 °C and the UV-detector was set at λ_{max} 220 nm (Abdul Barry et al., 2006). The comparison was done to show the advantages of the developed UPLC method over the Rp-HPLC in terms of efficient and earlier separation (0.94 min), lower injection volume (1 μl), lower solvent consumption (0.5 ml/min), better linearity over a wider range of analyte concentrations (0.002–2.5 mg/ml), lower limit of detection (0.002 mg/ml) and higher correlation coefficient (r = 0.9997). The comparison is summarized in Table 5.

3.9. Application

The developed UPLC method has been successfully used for the quantification of DS concentration in drug content and dissolution studies of Voltaren® 75 mg in comparison to laboratory developed HPMC K15 M matrix tablets of DS. Fig. 9 shows an example of the dissolution profile of DS from both formulations in 900 ml pH 7.5 as stated in its individual monograph for extended release DS in USP XXXII. The results showed that the method has capability to quantify as low as 5% release of the drug.

Table 5 Comparison of the analytical data for the determination of diclofenac sodium between the developed Rp-UPLC and a previous Rp-HPLC method.

Analytical data	Rp-UPLC method	Rp-HPLC method
Injection sample volume	1 μl	20 μl
Flow rate	0.5 ml/min	1 ml/min
Detection limit (LOD)	2 μg/ml	3.4 μg/ml
Correlation coefficient (r)	0.9997	0.9945
Linear range	(0.002–2.5) mg/ml	(0.01–0.06) mg/ml

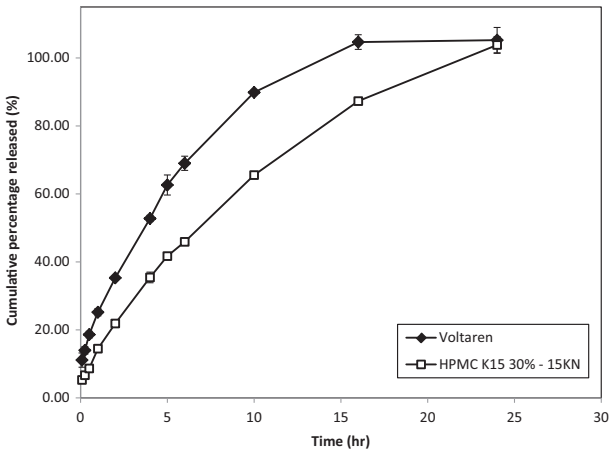

Figure 9 Release profile of DS from Voltaren® 75 mg and a matrix tablet in simulated intestinal medium (pH 7.5). Data are expressed as mean ± SD, n = 3.

4. Conclusion

A validated stability-indicating UPLC method has been developed for the analysis of DS in matrix formulations. It is shown above that the method was accurate, reproducible, rapid, linear, precise, and selective, proving the reliability of the method. The run time is short, i.e. 1.2 min, which enables rapid quantitation of many samples in routine analysis. No interference from the excipients was observed. The results demonstrated that the method would have a great value when applied in quality control and stability studies for DS.

Declaration of interest

The authors report no declaration of interest.

Acknowledgments

The authors would like to thank Al-Kayyali chair of pharmaceutical industry for hosting the work. Special thanks for TABUK pharmaceutical company, Tabuk, KSA for supplying us with DS. Also, thanks to Saudi FDA (SFDA) for supplying us with an authentic USP reference standard of DS.

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