

Eco-friendly Novel Kinetic Procedures for Spectrophotometric Analysis of Antihistamine Drug Part II. Validation, Practicality, and Green Perspective

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Abstract— A new kinetic technique to quantify desloratadine (DES) in dosage forms was interesting through the reaction of DES with 1,2-naphthoquinone-4-sulphonate (NQS), in an aqueous medium of borax at pH 9. The formed stable product was followed by measuring the increased absorbance with time at 485 nm. Four kinetic procedures were applied for the determination of DES. The initial rate and fixed-time (15 min) methods are the most suitable for plotting calibration graphs in the concentration ranges 2.50–35.00 and 2.50–37.50 µg/mL, with detection limits of 0.165 and 0.128 µg/mL, respectively. The molar absorptivity for the method was found to be 6.04×10^3 L/mol cm. The suggested kinetic methods were applied to determine DES in tablets. They were found to be more sustainable, eco-friendly, efficient, and practicable than the reported BP method, by applying green and white tools: AES, AGREE, AGREEprep, GAPI, and RGB, making it a safer alternative to be considered.

Keywords— Desloratadine, kinetic spectrophotometry, sodium 1,2-naphthoquinone-4-sulfonate (NQS), eco-friendly, tablets.

I. Introduction

Desloratadine is 8-chloro-11-(piperidin-4-ylidene)-6,11-dihydro-5H-benzo[5,6]cyclo-hepta[1,2-b]pyridine [1]. DES is a novel, non-sedating H1 receptor antagonist, with good tolerance and effective in the management of seasonal allergic rhinitis, perennial allergic rhinitis, and chronic urticaria [2]. A variety of methods have been reported to determine desloratadine in pharmaceutical preparations such as high-performance liquid chromatography (HPLC) [3–9], thin layer chromatography (TLC) [10,11], capillary zone electrophoresis [12], UV-spectrophotometry [13,14], spectrophotometry based on the formation of complexes with the chromogenic reagents [8,15–20], extractive spectrophotometry [21], spectrofluorimetry [8,22], voltammetry [23–25], and potentiometry [26]. Kinetic spectrophotometry was reported for the determination of desloratadine using acetaldehyde and 2,3,5,6-tetrachloro-1,4-benzoquinone (chloranil) in dioxan [27], and NQS in aqueous medium [28]. Most of the reported methods, including the kinetic method [27], need a lot of steps and organic solvents. While, kinetic spectrophotometry is the preferred choice for analytical chemists due to its high sensitivity, selectivity, and low detection limit [29,30].

Sodium 1,2-naphthoquinone-4-sulphonate (NQS) is a popular chromogenic reagent used for the spectrophotometric determination of many pharmaceutical primary and secondary amines due to its efficient reactivity and high reaction rate [31–33]. Therefore, this work aimed to improve eco-friendly novel kinetic procedures to determine desloratadine spectrophotometrically based on the reaction with NQS in an aqueous medium.

Multiple tools in green analytical chemistry (GAC) are used to evaluate the greenness of analytical performance and the environmental impact of analytical procedures, such as AGREEprep, the Analytical Eco-Scale (AES), Analytical Greenness (AGREE), and the Green Analytical Procedure Index (GAPI). For the assessment of analytical sample preparation processes' greenness and their environmental impact, the AGREEprep tool is used. The AGREEprep tool includes ten criteria covering different aspects that contribute to the overall sample preparation greenness. The efficiency of each section is evaluated with a score ranging from 0 to 1, where 1 is the ideal level of performance [34,35]. The AES scale is based on evaluating penalty points related to the number of reagents or solvents, the amounts of

hazardous materials consumed, and the waste generated during the analytical procedures. The total of these points is calculated and subtracted from 100. A total score of >75 indicates that the analytical method has excellent greenness; a total score ranging from 50 to 75 indicates acceptable greenness. Finally, a total score of <50 indicates insufficient greenness [36,37]. The AGREE tool created a twelve-segment pictogram, representing all the principles of green analytical chemistry. Typically, a score of 1 is given for green analytical procedures, with all-green twelve segments; variation from typical green criteria leads to lower scores and lower color scales (bottle green, orange, yellow, and red) [38]. The Green Analytical Procedure Index (GAPI) is a fifteen-segment pictogram representing different aspects of the analytical process from sampling to detection [39]. White analytical chemistry (WAC) is an expanded version of the GAC methodology, referring to the red-green-blue color model of light, where red indicates the quality of the analytical process, such as accuracy and sensitivity. Blue suggests the practicality and economics of the analytical method as confirmed by cost estimation, time efficiency, other requirements, and operational simplicity. The overall quality of the analytical process is expressed through whiteness. A method is not necessarily greener if it is whiter and more comprehensive. The RGB model is based on the twelve principles of WAC that help in assessing the sustainability of an analytical method. RGB analysis is a semi-quantitative approach that classifies analytical methods into three color-coded groups based on their environmental impact [40,41].

II. Materials and methods

2.1. Instrumentation

UV–VIS spectrophotometer V–530 (Jasco, Japan), 1 cm quartz cells, and circulating water to keep the temperature of the cell constant were used for the absorbance measurements of samples. Spectra were automatically obtained by the Jasco system software. A pH meter Consort C830 (Belgium) equipped with a combined glass pH electrode and a Thornton sonicator (model T–14) was also used.

2.2. Chemicals and pharmaceutical samples

All chemicals used were of analytical reagent grade. Sodium 1,2-naphthoquinone-4-sulphonate (NQS) from Aldrich Chemical Co., St. Louis (USA), borax, $\text{Na}_2\text{B}_4\text{O}_5(\text{OH})_4 \cdot 8\text{H}_2\text{O}$ = 381.36 g/mol, and methanol were purchased from BDH (England) and Merck (Germany), respectively. Doubly distilled water was used in all experiments. Working reference standard of desloratadine (DES), $\text{C}_{19}\text{H}_{19}\text{ClN}_2$ = 310.82 g/mol, and bulk DES powder samples were supplied by CADILA Healthcare Limited (India); their purity was 99.94% and 99.83%, respectively, according to the BP method [1]. Newloratadine and Desrinal tablets are labeled to contain 5 mg desloratadine per tablet, and are purchased from the local market.

2.3. Solutions

Aqueous solution of 0.2% w/v NQS was prepared by dissolving the appropriate weight of NQS in water. The solution was freshly prepared and protected from light during use. Borax solution 5×10^{-3} mol/L was prepared by dissolving the appropriate weight of borax in water and adjusting to pH 9.0.

2.4. Standard solutions of DES

A standard stock solution of DES was prepared by dissolving the appropriate weight of the working reference standard DES in methanol to obtain 0.5 mg/mL. This stock solution was further diluted with water to obtain working solutions in the range of 2.50–37.50 µg/mL.

2.5. Sample solutions

A 0.5 mg/mL stock solution of DES was prepared by dissolving the appropriate weight of the bulk DES powder in methanol, sonicating for 5 min, and filtering. Different diluted solutions in water were prepared and tested using the kinetic procedures and BP method [1].

2.6. Commercial formulation solutions

Twenty tablets, each containing 5 mg of DES, were accurately weighed and powdered. An accurately weighed portion of the powder equivalent to about 12.5 mg of DES was transferred into a 25 mL calibrated flask, and 15 mL of methanol was added. The solutions were sonicated for 10 min and completed to volume with methanol to achieve a 0.5 mg/mL concentration of DES. The contents were mixed well and filtered. To study the excipient's disability (if any), about 25 mg of placebo (desloratadine excipient) was accurately weighed into a 25 mL volumetric flask then the same procedures were followed to dilute the tablets. The spectrum of the placebo shows no peak in the range of 400–700 nm.

2.7. Recommended kinetic procedures for the determination of DES

2.7.1. Initial rate method

Aliquots of 2.50–37.50 µg/mL of standard DES solution (0.05–0.75 mL, 0.5 mg/mL) were transferred into a series of 10 mL calibrated volumetric flasks. In each flask, 1.25 mL of 0.2% NQS solution and 0.2 mL of borax solution (pH 9.0) were added. The volume was made up to the mark with distilled water at 25 ± 1 °C. The contents of each flask were mixed well, and the increase in absorbance was recorded at 485 nm as a function of time between 0 and 35 minutes against a reagent blank treated similarly. The initial rate of the reaction (v) at different concentrations was obtained from the slope of the tangent to the absorbance-time curve. The calibration curve was constructed by plotting the logarithm of the initial rate ($\log v$) vs the logarithm of the molar concentration of the DES ($\log C$). The amount of the drug was obtained either from the calibration graphs or the regression equation.

2.7.2. fixed time method

The same series of selected standard concentrations was prepared, and the absorbance of these solutions was measured at 485 nm after 900 s against a reagent blank. The absorbance plotted against the DES concentration gave the calibration graph. The DES content in each sample was evaluated using the regression equation.

2.8. Kinetic methods validation

The proposed kinetic spectrophotometric methods were validated following the guidelines provided by the International Conference on Harmonization (ICH), specifically the Q2 (R2) guidelines [42].

2.8.1. Linearity

The calibration graph was constructed by applying the general assay procedure using the initial rate and fixed time methods for DES determination.

2.8.2. Limits of detection and quantification

The limits of detection (LOD) and quantitation (LOQ) were determined experimentally from regression equations of DES. The LOD was expressed as the concentration of drug that generated a response three times the signal-to-noise (S/N) ratio, and the LOQ was 10 times the S/N ratio. The LOD of DES attained as defined by IUPAC [43]; $LOD_{(k=3.3)} = k \times S_a / b$, $LOQ_{(k=10)} = k \times S_a / b$ (where S_a is the standard deviation of the intercept and b is the slope of the calibration curve).

2.8.3. Precision and accuracy

DES samples with different concentrations (2.5, 10, 20, and 35 $\mu\text{g/mL}$) were tested five times on the same day and measured to estimate the intraday precision.

The interday precision was evaluated by testing the same prepared samples of DES on three different days. The relative standard deviation (RSD) of the assay values was calculated. The accuracy was estimated for the same four different concentrations. The mean recovery of DES was calculated from the corresponding linear regression equation by applying the following relation.

$$\text{Recovery \%} = C_{\text{found}} / C_{\text{taken}} \times 100\% \quad (1)$$

2.8.4. Robustness

The robustness of an analytical procedure is usually studied to assess its ability to remain unaffected when small changes are applied to the method's approved parameters. Changes have included analyzing the effect of changing the NQS, borax volumes, and measurement time.

III. Results and discussion

In our previous work [28], NQS has been used for the kinetic spectrophotometric determination of DES by applying the fixed time procedure. To generate the nucleophile from DES and activate the nucleophilic substitution reaction, an alkaline medium was necessary, since the results revealed that DES does not react with NQS in acidic media. The reaction of NQS with DES in an alkaline medium at 25 °C was resulted in the formation of an intense orange-colored product that exhibits an absorption maximum at 485 nm (Fig. 1). The pure drug and the reagent blank (NQS) showed a negligible absorbance at the corresponding maximum at 485 nm.

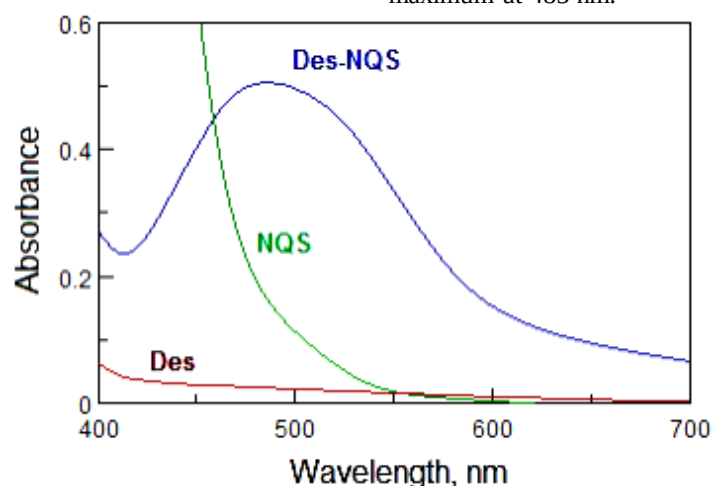


Figure 1. Absorption spectra of (a) DES solution (25 $\mu\text{g/mL}$) against methanol, (b) reagent blank against water, and (c) DES–NQS system against reagent blank ($C_{\text{DES}}=25 \mu\text{g/mL}+1.25 \text{ mL}$ of 0.2% NQS+0.2 mL of borax $5 \times 10^{-3} \text{ M}$ in 10 mL final volume); reaction time: 15 min.

Several parameters such as reagent concentrations, buffer, time, temperature, and sequence of additions have been optimized to achieve high sensitivity, low blank reading and high stability for the determination of DES. Best results were obtained in the borax buffer solution $5 \times 10^{-3} \text{ M}$, pH 9.0. Also, it has been found that the absorbance of the reaction solution increased as the NQS concentration increased, and the highest

absorption intensity was attained when the volume of NQS was 1.25 mL of 0.2% (w/v). Increasing the volume of NQS leads to a decrease in the absorbance; this may be due to the high background absorbance of the reagent (Fig. 2). Study for optimization of borax concentration revealed that the optimum volume of borax was 0.20 mL of $5 \times 10^{-3} \text{ M}$ (Fig. 2). At this value, the amino group of DES facilitates the nucleophilic

substitution reaction. At higher concentrations of borax, the absorbance of the solution decreased. This was probably attributed to the increase in the amount

of hydroxide ion that holds back the condensation reaction between DES and NQS.

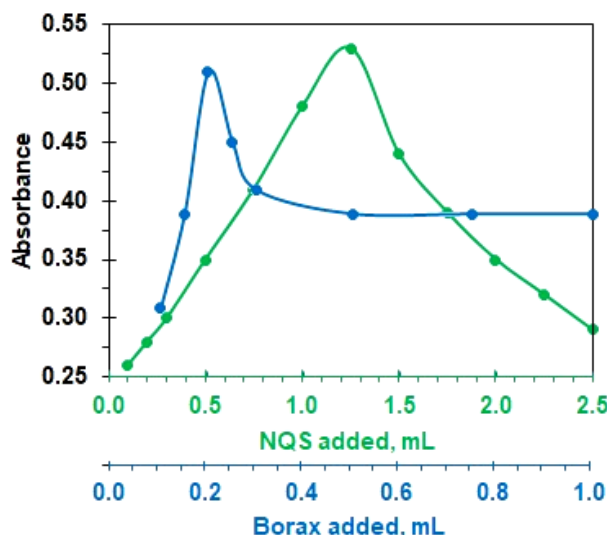


Figure 2. Effect of volume of NQS 0.2% w/v in the presence of 0.2 mL of borax 5×10^{-3} M and borax 5×10^{-3} M in the presence of 1.25 mL of NQS 0.2% on the formation of colored product DES (25 $\mu\text{g/mL}$)–NQS, temperature: 25 ± 0.5 °C; reaction time: 15 min.

The reaction rate of DES with NQS at pH 9.0 slightly increased with increasing temperature in the range of 15 to 45 °C; increasing the temperature had a negative effect on the absorption values of the reaction. This was probably attributed to the instability of the DES–NQS derivative. The optimum temperature for the determination process was selected at 25 °C. The equilibrium of the reaction appears after 15 minutes, and the color remains stable for 24 hours, so this condition was exploited to determine the DES content kinetically.

3.1. Stoichiometry

The limiting logarithmic method [44] was employed to evaluate the stoichiometry of the DES–NQS complex. Two sets of solutions were prepared; the concentration of DES was constant in the first set, while the

concentration of NQS was constant in the second. The absorbance was measured for each series at 485 nm, log absorbance versus log C_{NQS} or log C_{DES} was plotted, and the slope of the respective line was estimated (Fig. 3a and b). Finally, the slope of the DES line (1.0711) was divided over a slope of the NQS line (0.9618) the ratio of the reaction was found to be 1:1 DES to NQS. We also found from Job's method of continuous variation and mole-ratio that a molar ratio is 1:1 DES to NQS, with the formation constant (log K_f) value of DES–NQS complex is 6.10 at the ratio 1:1 [28].

Based on the observation stoichiometric ratio and the presence of only one center (N–H group) in the DES molecule that is available for the substitution reaction, the reaction pathway was postulated to proceed as shown in Scheme 1.

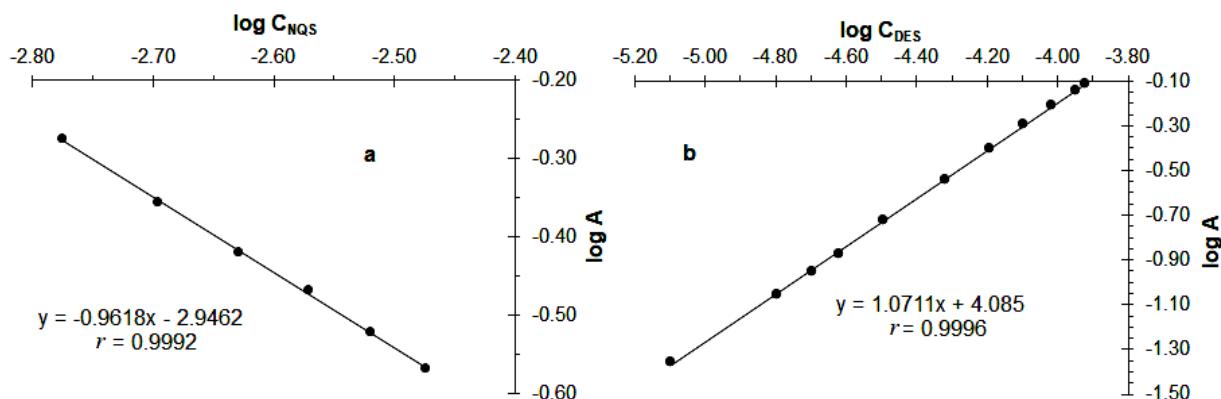
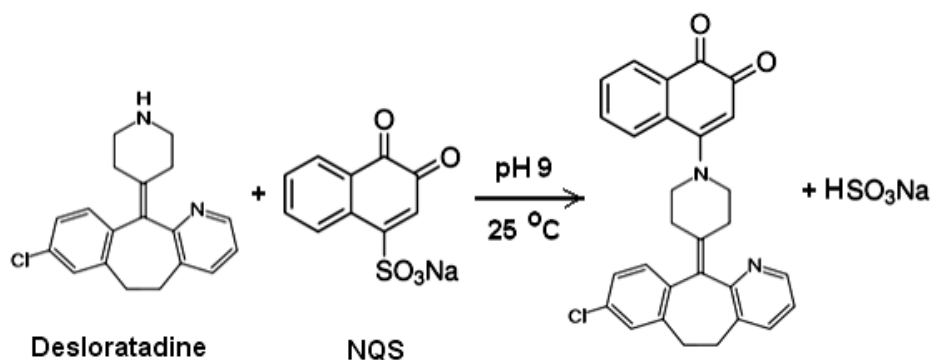


Figure 3. Stoichiometry of the reaction between DES and NQS adopting the limiting logarithmic method. (a) $C_{\text{DES}} = 8.043 \times 10^{-5}$ M, $C_{\text{NQS}} = 1.68 \times 10^{-3}$ – 3.36×10^{-3} M and (b) $C_{\text{NQS}} = 9.6 \times 10^{-3}$ M, $C_{\text{DES}} = 8.043 \times 10^{-6}$ – 1.206×10^{-4} M.



Scheme 1. Structure of DES and its reaction product with NQS.

3.2. Kinetic analytical data

Under the above-described optimum conditions, the absorbance-time curves for the reactions of DES with NQS in alkaline medium were generated (Fig. 4). This was used as the basis for a useful kinetic method for

the determination of DES. The initial rate, rate constant, variable time (fixed concentration or fixed absorbance), and fixed time methods were tested, and the most suitable analytical methods were chosen regarding the applicability, sensitivity, and the values of the intercept and correlation coefficient (r).

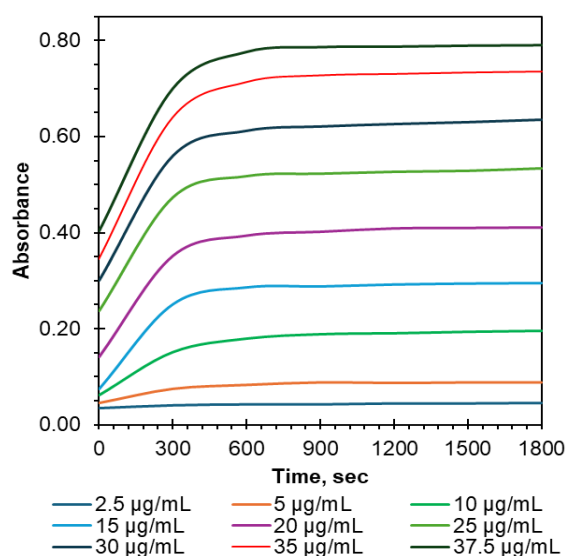


Figure 4. Absorbance-time curve at 485 nm for the reaction of DES with NQS in borax buffer at pH 9; C_{DES} = 2.5–37.5 $\mu\text{g/mL}$.

3.2.1. Initial rate method

The initial rate of reaction would follow a pseudo-order rate constant and obey the following rate equation:

$$\text{Rate } (v) = \Delta A / \Delta t = k [C]^n$$

where A , t , k , C , and n are absorbance, measuring time, pseudo-order rate constant, the concentration of DES (mol/L), and order of the reaction (slope), respectively. The logarithmic form of the above equation is given as follows:

$$\log v = n \log C + \log k$$

A calibration curve was constructed by plotting the logarithm of the initial rate of reaction ($\log v$) vs $\log C_{DES}$, which showed a linear relationship over the concentration range of 8.04×10^{-6} – 11.26×10^{-5} mol/L (2.5–35.0 $\mu\text{g/mL}$) with $r=0.9998$, as presented in Fig. 5 and equation (2).

$$\log v = 1.1292 \log C_{DES} + 0.7947 \quad (2)$$

The slope value of equation (2) is $n=1.1292$ (≈ 1.0), describing the first order of the reaction. Thus, $\log k = 0.7947$ and then $k=6.23 \text{ S}^{-1}$

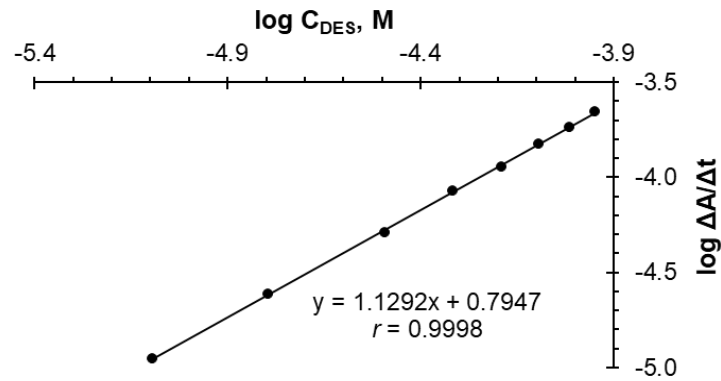


Figure 5. Calibration plot of the logarithm of the reaction rate against the logarithm of the molar concentration of DES for the initial rate method.

3.2.2. Rate constant method

Graphs of log absorbance versus time within the DES concentration range of 2.5–37.5 $\mu\text{g/mL}$ were constructed. Pseudo order rate constant (k) corresponding to different DES concentrations was calculated from the slopes multiplied by -2.303 (Table 3)

1). The regression equation of C vs k was found to be linear (Fig. 6) over the range 10.0–35.0 $\mu\text{g/mL}$ (3.21×10^{-5} – 11.26×10^{-5} mol/L) with $r = 0.9857$ (equation 3).

Table 1. Values of rate constant k for different rates of variable concentration of DES.

C_{DES} (mol/L)	k (1/S)
3.21×10^{-5}	-10.13×10^{-4}
4.82×10^{-5}	-8.75×10^{-4}
6.43×10^{-5}	-8.06×10^{-4}
8.04×10^{-5}	-6.91×10^{-4}
9.65×10^{-5}	-6.21×10^{-4}
11.26×10^{-5}	-5.52×10^{-4}

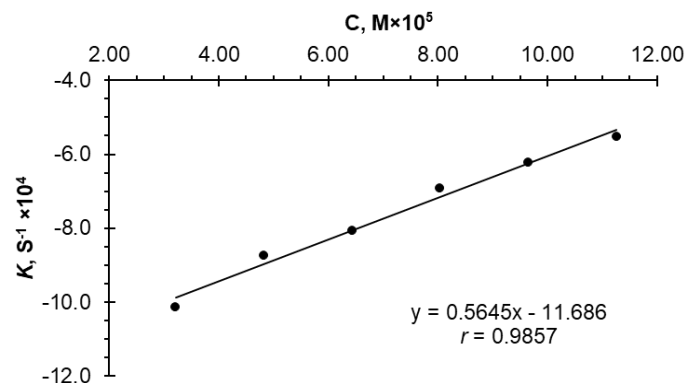


Figure 6. Calibration plot of DES for the rate constant method.

3.2.3. Fixed absorbance method

Time values in seconds were measured for different DES concentrations at a specific absorbance value of 0.40 chosen in Fig. 4. The reciprocal of time ($1/t$) was calculated (Table 2), a graph of the relationship of $1/t$ to DES concentration was plotted (Fig. 7). The regression equation (4) was found to be linear in the range 20.0–35.0 $\mu\text{g/mL}$ (6.43×10^{-5} – 11.26×10^{-5} mol/L), with $r = 0.9946$.

to DES concentration was plotted (Fig. 7). The regression equation (4) was found to be linear in the range 20.0–35.0 $\mu\text{g/mL}$ (6.43×10^{-5} – 11.26×10^{-5} mol/L), with $r = 0.9946$.

Table 2. Reciprocal time is taken at fixed absorbance (0.40) for the different rates of variable concentration of DES. NQS concentration is constant.

C_{DES} (mol/L)	$1/t$ (S^{-1})
6.43×10^{-5}	16.66×10^{-4}
8.04×10^{-5}	55.55×10^{-4}
9.65×10^{-5}	100.00×10^{-4}
11.26×10^{-5}	153.84×10^{-4}

3.2.4. Fixed time method

Depending on Fig. 4, the absorbance values of the complex solutions formed using variable concentrations of DES were recorded at specific times.

The calibration graph of absorbance versus DES concentration at each fixed time was constructed and

then the linear regression equations and correlation coefficients of these calibration curves were obtained [45], as presented in Table 3. This shows that 15 min is the optimal time to achieve the most acceptable linearity for evaluation of DES

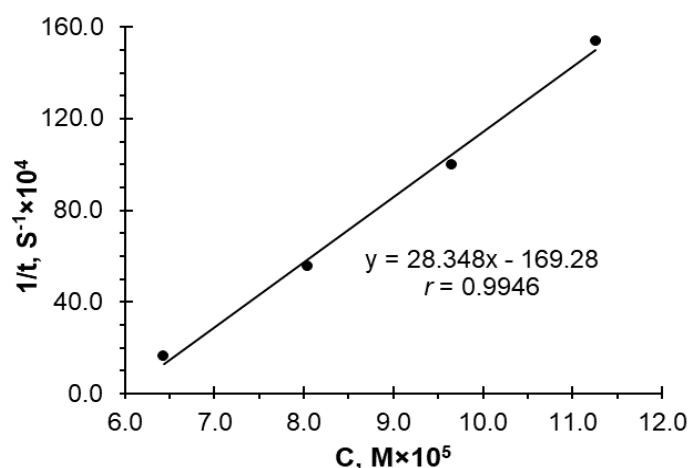


Figure 7. Calibration plot of DES for the fixed absorbance method.

3.3. Validation of the proposed methods

3.3.1. Linearity, LOD and LOQ

By comparing the studied kinetic procedures, it was found that the most acceptable values of the correlation coefficient and more reaction products (indicated by higher absorbance readings) were obtained for the initial rate and fixed time (15 min) methods which were chosen for the validation study. Linearity of the initial rate method was confirmed over the concentration range of 2.5–35.0 µg/mL DES with

LOD and LOQ of 0.165 µg/mL and 0.502 µg/mL, respectively. Table 4 shows the calibration data and statistical parameters of the fixed-time method. The proposed method has excellent sensitivity through low values of both LOD and LOQ.

3.3.2. Accuracy and precision

The accuracy and precision of the proposed methods were established through their application in the estimation of four different standard solutions of DES (2.5, 10, 20, and 35 µg/mL). Intra and inter-day assay variation was evaluated by measuring these standard solutions on the same day and three different days from day 1 to 8 after preparation. Mean percentage recoveries ranged from 100.36 to 101.05 %. This close agreement of our results to the true values is indicative of the high accuracy. The RSD% of the assay values was calculated in view to justify the precision. Low values of RSD% (<2 %) indicate the high precision of the developed method (Table 5).

Table 3. Regression equations for DES at different fixed times and 25 °C.

Time (min)	Regression equation	Correlation coefficient (r)	Linear range (µg/mL)
0	A = 0.0153C–0.0100	0.9994	2.50–37.50
5	A = 0.0194C–0.0178	0.9994	2.50–37.50
10	A = 0.0206C–0.0196	0.9996	2.50–37.50
15	A = 0.0209C–0.0177	0.9999	2.50–37.50
20	A = 0.0213C–0.0185	0.9997	2.50–37.50
25	A = 0.0215C–0.0194	0.9995	2.50–37.50
30	A = 0.0216C–0.0191	0.9993	2.50–37.50

A, absorbance; C, concentration.

Table 4. Analytical characteristics of the fixed time (15 min) method.

Parameters	DES
$\lambda_{\max}$ (nm)	485
Linear range ($\mu\text{g/mL}$)	2.50–37.50
Intercept (a)	–0.0177
Standard deviation of the intercept (S_a)	8.13×10^{-4}
Slope (b)	0.0209
Standard deviation of the slope (S_b)	4.21×10^{-4}
Standard deviation of residuals ($S_{x/y}$)	5.42×10^{-3}
RSD% of the slope	2.01
Correlation coefficient (r)	0.9999
Ringbom optimum concentration range ($\mu\text{g/mL}$)	7.50–35.00
LOD ($\mu\text{g/mL}$)	0.128
LOQ ($\mu\text{g/mL}$)	0.389
Molar absorptivity (L/mol cm)	6.04×10^3
Stability (hrs)	24
Logarithmic formation constant	6.10
Sandell's sensitivity ($\mu\text{g/cm}^2$ per 0.001 absorbance unit)	0.022
Mean recovery $\pm$ SD%	100.67 $\pm$ 0.29

Table 5. Accuracy and precision for the determination of DES reference standard by the proposed kinetic methods.

Taken $\mu\text{g/mL}$	Initial rate method				Fixed time method			
	Found $\pm$ SD $\mu\text{g/mL}$	Er %	RSD %	Recovery %	Found $\pm$ SD $\mu\text{g/mL}$	Er %	RSD %	Recovery %
2.50	2.51 $\pm$ 0.04	0.40	1.59	100.40	2.51 $\pm$ 0.05	0.40	1.99	100.50
10.00	10.09 $\pm$ 0.07	0.90	0.64	100.90	10.08 $\pm$ 0.04	0.80	0.40	100.80
20.00	20.21 $\pm$ 0.07	1.05	0.35	101.05	20.20 $\pm$ 0.08	1.00	0.39	101.02
35.00	35.25 $\pm$ 0.09	0.71	0.25	100.71	35.13 $\pm$ 0.07	0.36	0.20	100.36
Mean $\pm$ SD				100.76 $\pm$ 0.28				100.67 $\pm$ 0.29
N				5				5
RSD%				0.277				0.288
SAE				0.125				0.129

* SD: Standard deviation; N: number of experiments; RSD: Relative standard deviation; SAE: Standard analytical error.

3.3.3. Robustness

In the current study, the robustness of the suggested method was ascertained by variation in the volume of 0.2% (w/v) NQS (± 0.10 mL), 5×10^{-3} mol/L borax (± 0.02 mL), and measurement time (± 5 min). Under the varied conditions sample solutions of DES were analyzed at a concentration level of 25.0 $\mu\text{g/mL}$. Results prove that the proposed methods are robust and acceptable with excellent recoveries (Table 6).

Table 6. Results of robustness study of DES.

Parameter variation	Recovery % $\pm$ SD ^a	
	Initial rate	Fixed time
No Change	100.76 $\pm$ 0.28	100.67 $\pm$ 0.29
Change in NQS (0.2% w/v) volume		
1.15 mL	98.86 $\pm$ 0.22	98.88 $\pm$ 0.21
1.35 mL	99.82 $\pm$ 0.26	99.85 $\pm$ 0.17
Change in borax 5×10^{-3} M volume		
0.18 mL	98.96 $\pm$ 0.20	99.83 $\pm$ 0.19
0.22 mL	98.88 $\pm$ 0.18	98.93 $\pm$ 0.22
Change in measurement time		
10 min	100.03 $\pm$ 0.19	100.08 $\pm$ 0.18
20 min	100.06 $\pm$ 0.25	100.12 $\pm$ 0.20

^aAverage of five determinations.

3.5. Application in dosage forms

DES in marketed formulations was determined by applying the optimized kinetic procedures. The

3.4. Application to bulk powder

The proposed kinetic spectrophotometric and reported BP HPLC [1] methods were applied to determine DES in bulk powder samples. To prepare the sample solution of DES, a definite amount of bulk powder was dissolved in methanol. By applying the developed kinetic method, four levels of DES concentrations (2.5, 10, 20, and 35 $\mu\text{g/mL}$) were prepared and analyzed under the same optimum conditions. The established results are given in Table 7.

recovery results for the assay DES (Table 8) were compared with those of the official method [1]. The commonly used excipients and additives in these

formulations did not interfere with the analysis. The kinetic developed method was compared with the reference method by calculation of Student's *t*- and *F*-tests at 95% confidence limit for five degrees of freedom [45]. The established *t*- and *F*-values do not exceed the theoretical ones, indicating no significant differences between the two methods. The recovery results showed that the improved method has high efficiency for the routine analysis of DES in pharmaceutical preparations.

The feasibility of the current testing method in the field of pharmaceutical quality control is clearly demonstrated by its success when applied to the analysis of raw material samples and commercial pharmaceutical preparations. Simply substitute the absorbance value into the linear regression equation for the reference standard and then quickly calculate the concentration.

Table 7. Application of the proposed and BP methods for the determination of DES in bulk powder.

Conc. taken (µg/mL)	Initial rate method	Fixed time method	BP HPLC method [1]
	Found ^a %	Found ^a %	Found ^a %
2.50	99.83	99.85	99.74
10.00	99.81	99.86	99.79
20.00	99.90	99.89	99.95
35.00	99.82	99.82	99.83
Mean±SD	99.84±0.041	99.85±0.029	99.83±0.089
RSD %	0.041	0.029	0.089
Variance	1.25×10 ⁻³	6.50×10 ⁻⁴	6.02×10 ⁻³
<i>t</i> -test	2.24*	2.25*	2.23*
<i>F</i> -value	1.01*	1.02*	

^a The average of 5 separate results. *The tabulated *t* and *F* values were 2.776, 6.26, respectively at *P* = 0.05.

3.6. Greenness and whiteness assessment of the suggested method

We investigated the proposed method for the assay of DES in its tablets using ASE, AGREE, AGREEprep, and GAPI tools, to emphasize the importance of green analysis of pharmaceutical drugs. The Analytical Environmental Assessment (AES) criteria are detailed in Table 9, where penalty points are calculated based on the method's adverse environmental impacts. The overall Analytical Environmental Score shows that the proposed kinetic method achieves the highest environmental score compared to the BP method, which consumes more energy and produces a greater quantity of solvents and waste. Table 10 demonstrates the greenness and whiteness profiles of the proposed method using the mentioned metrics and its

comparison with the reported formal HPLC. The practical application of AGREEprep showed a value of 0.75 for our method. The greenness of the sample preparation of the proposed method was compared with that of the previously mentioned method in BP for DES determination. The proposed method was found to be greener and safer. Using AGREE, our method showed a score (0.87) that ranked first, ahead of the formal BP HPLC method. This distinctive visual indicator confirms the eco-friendly features of the recommended methodology. In conclusion, the application of the AGREE approach reinforces the idea that the proposed method is well aligned with the principles of GAC, enhancing the AGREE toolkit that uses the environment.

Table 8. Determination of DES in dosage forms by the proposed kinetic and BP methods.

Sample	Taken (µg/mL)	Fixed time method		Initial rate method		BP method [1]	
		Found (µg/mL)	Recovery (%)	Found (µg/mL)	Recovery (%)	Found (µg/mL)	Recovery (%)
Desrinal (5 mg/tablet) (Bioframa, Türkiye)	5.00	4.99	99.82	4.99	99.83	5.01	99.90
	10.00	10.01	100.10	9.99	99.91	9.97	99.67
	30.00	29.96	99.87	30.01	100.03	29.97	99.92
	Mean±SD	99.93±0.15		99.92±0.10		99.83±0.14	
	N	5		5		5	
	RSD%	0.15		0.10		0.27	
	<i>t</i> -test	1.15		1.79		2.71	
	<i>F</i> -test	0.87		1.96		-	
Newloratadine (5mg/tablet) (BPI, Syria)	5.00	4.98	99.00	4.94	98.80	4.93	98.60
	10.00	9.98	99.80	9.97	99.70	9.94	99.40
	30.00	30.03	100.10	29.99	99.98	30.02	100.07
	Mean±SD	99.63±0.57		99.50±0.62		99.36±0.74	
	N	5		5		5	
	RSD%	0.22		0.62		0.74	
	<i>t</i> -test	1.63		1.80		1.93	
	<i>F</i> -test	1.45		1.42		-	

^aFive independent analyses. Theoretical values for *t* and *F*-values at five degrees of freedom and 95% confidence limit are *t* = 2.776 and *F* = 6.26.

Table 9. Penalty points (PPs) of the proposed kinetic spectrophotometric and BP methods according to the Analytical Eco-Scale to determine DES.

Evaluation Points	Penalty Points (PPs)	
	The proposed method	BP HPLC method [1]
Reagents		
Methanol	6	-
NQS 0.2% (w/v)	0	-
Borax buffer	2	-
Acetonitrile	-	8
Sodium dodecyl sulfate	-	2
Trifluoroacetic acid	-	4
Instrument		
Energy consumption	0 (≤ 0.1 kWh per sample)	1 (≤ 1.5 kWh per sample)
Waste generation	1 (< 1 mL (g))	5 (> 10 mL (g))
Waste treatment	3	3
Occupational hazard	0	0
Total PPs	$\Sigma 11$	$\Sigma 23$
Analytical Eco-Scale score (100-pp)	88	77
Assessment	Excellent greenness	Acceptable greenness

The GAPI index indicates that the proposed method exhibits three yellow steps in its GAPI index, with one red-colored step indicating the area of no treatment of waste, while the result revealed the greenness of most. So, this newly developed method is considered to be eco-friendly. On the other hand, the HPLC method approved by the British Pharmacopoeias obtains fewer green and more yellow steps, but it has two red-colored steps, suggesting potential environmental drawbacks.

The superiority of the recommended green methodology is due to the shortened sample preparation and analysis stages, speed of analysis, and the absence of hazardous wastes in the proposed method, in addition to energy savings. While the BP method uses toxic solvents such as acetonitrile in large quantities in addition to some chemicals, which results in untreated wastes, it also consumes more energy due to heating the column oven to 35 °C. All applied greenness assessment tools have demonstrated a significant potential for environmental harm from pharmacopoeial technologies.

The whiteness of the analytical process was measured using the RGB model algorithm. The red color included the performance efficiency of the analytical

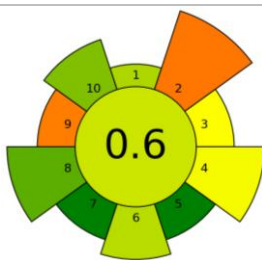
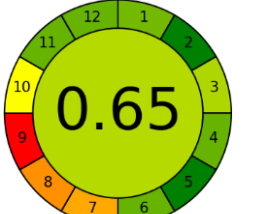
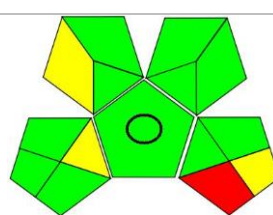
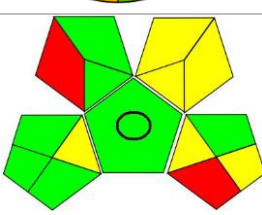
method, such as the application range, LOD, LOQ, accuracy, and precision. The green color included the eco-friendly parameters, such as energy consumption, direct effect of solvents/reagents, their consumption, and toxicity.

The blue color included the productivity parameters of the analytical method, such as operational simplicity, cost, and time efficiency, as represented in Table 10. The developed kinetic method achieved the highest red score of 97.5 for analytical performance, while the BP method achieved a lower red score of 93.7 due to the higher detection level and quality level.

WAC, as we noted in the introduction, allows for the formal and systematic expression of all the main expectations regarding the required method features, including green and other residual requirements.

The overall result of the proposed kinetic and BP techniques, which is the average result of the evaluation of all twelve principles - whiteness (%), is shown in Table 10.

Table 10. Assessment of greenness and whiteness and comparative study of the proposed method with BP method for determining the amount of DES in dosage forms.

Assessment tool	Proposed kinetic method	BP HPLC method [1]																																																																																
AES	88	77																																																																																
AGREEprep																																																																																		
AGREE																																																																																		
GAPI																																																																																		
RGB	<table><tr><th colspan="4">Method: Proposed CGC-FID method</th></tr><tr><td>R1: Scope of application</td><td>100.0</td><td>G1: Toxicity of reagents</td><td>76.0</td><td>B1: Cost-efficiency</td><td>100.0</td></tr><tr><td>R2: LOD and LOQ</td><td>90.0</td><td>G2: Amount of reagents and waste</td><td>100.0</td><td>B2: Time-efficiency</td><td>110.0</td></tr><tr><td>R3: Precision</td><td>100.0</td><td>G3: Energy and other media</td><td>100.0</td><td>B3: Requirements</td><td>100.0</td></tr><tr><td>R4: Accuracy</td><td>100.0</td><td>G4: Direct impacts</td><td>98.3</td><td>B4: Operational simplicity</td><td>63.3</td></tr><tr><td></td><td>97.5</td><td>93.6</td><td></td><td>93.3</td><td></td></tr><tr><td></td><td colspan="4">94.8</td><td></td></tr></table>	Method: Proposed CGC-FID method				R1: Scope of application	100.0	G1: Toxicity of reagents	76.0	B1: Cost-efficiency	100.0	R2: LOD and LOQ	90.0	G2: Amount of reagents and waste	100.0	B2: Time-efficiency	110.0	R3: Precision	100.0	G3: Energy and other media	100.0	B3: Requirements	100.0	R4: Accuracy	100.0	G4: Direct impacts	98.3	B4: Operational simplicity	63.3		97.5	93.6		93.3			94.8					<table><tr><th colspan="4">Method: BP-HPLC method</th></tr><tr><td>R1: Scope of application</td><td>90.0</td><td>G1: Toxicity of reagents</td><td>80.0</td><td>B1: Cost-efficiency</td><td>100.0</td></tr><tr><td>R2: LOD and LOQ</td><td>85.0</td><td>G2: Amount of reagents and waste</td><td>70.0</td><td>B2: Time-efficiency</td><td>100.0</td></tr><tr><td>R3: Precision</td><td>100.0</td><td>G3: Energy and other media</td><td>90.0</td><td>B3: Requirements</td><td>90.0</td></tr><tr><td>R4: Accuracy</td><td>99.9</td><td>G4: Direct impacts</td><td>96.7</td><td>B4: Operational simplicity</td><td>60.0</td></tr><tr><td></td><td>93.7</td><td>84.2</td><td></td><td>87.5</td><td></td></tr><tr><td></td><td colspan="4">88.5</td><td></td></tr></table>	Method: BP-HPLC method				R1: Scope of application	90.0	G1: Toxicity of reagents	80.0	B1: Cost-efficiency	100.0	R2: LOD and LOQ	85.0	G2: Amount of reagents and waste	70.0	B2: Time-efficiency	100.0	R3: Precision	100.0	G3: Energy and other media	90.0	B3: Requirements	90.0	R4: Accuracy	99.9	G4: Direct impacts	96.7	B4: Operational simplicity	60.0		93.7	84.2		87.5			88.5				
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IV. Conclusion

Developing suitable analytical methods is crucial for monitoring the quality of medicines, a goal pursued by reputable pharmaceutical companies to achieve proven safety, quality, and efficacy. Therefore, this work presents an eco-friendly, efficient, and reliable kinetic spectrophotometric method for determining the concentration of the antihistamine desloratadine (DES) based on the coupling reaction of DES with NQS in both pure form and in tablets, utilizing a simple sample preparation step in methanol. It is a straightforward and robust method and is more accurate and sensitive than similar reported methods with LOD 0.128 µg/mL. This eco-friendly method was evaluated, and purity and whiteness assessment criteria (AES, AGREEprep, AGREE, GAPI, BAGI, and RGB12) demonstrated that the developed method is eco-friendly, white, applicable, and efficient based on all obtained data. It surpasses the analytical method used in the BP due to its simplicity, the absence of environmentally polluting chemicals, and its lower

cost. Furthermore, this method does not generate hazardous waste that poses a risk to public health and the environment. Consequently, the proposed method is highly suitable for monitoring the quality of DES in tablets.

Declaration of Conflicting Interests

The author declares that he has no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Human Participants

This paper does not involve human participants.

Originality Note

The author confirms that the manuscript is his original work, and

if others' works are used, they are properly cited/quoted.

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About the author

Prof. Safwan Ashour is started at Aleppo University in 1994, where he was promoted to Associate Professor in 1998 and Full Professor in 2005. In 2002, he joined the Environmental Research Center at Brunel University, London, UK, as a research fellow with a grant awarded by the Syrian Ministry of Higher Education. Prof. Ashour previously taught analytical chemistry and analytical Biochemistry courses in

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Research 2010 (Syria); Served as a reviewer for more than 48 international journals specialized in analytical chemistry and its applications; The number of Editorial Appointments in Journals/ Conferences (Editor, Reviewer and Member) between 2004 and 2025 is 129. No. Invited Speaker/ Resource person, between 2019 and 2025, is 229. According to Google Scholar, his citations are 812, and his H-index is 18.



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