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Research Article
Titrimetric and Spectrophotometric Assay of Diethylcarbamazine Citrate in Pharmaceuticals Using N-bromosuccinimide

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Abstract

Background: Diethylcarbamazine citrate (DEC) is the drug of choice for the treatment of lymphatic filariasis, a disease which affects people in the tropical areas of the world. Mass treatment with DEC is recommended for control of lymphatic filariasis and in this connection, there is a need for an assay which could be used for the purpose of medication control. **Objective:** The present study is aimed at developing titrimetric and spectrophotometric methods for the determination of DEC in bulk as well as in dosage forms, using N-bromosuccinimide (NBS). **Materials and Methods:** In both methods, DEC is treated with a measured excess of standard NBS in HCl medium and after a standing time of 15 min, the residual NBS is determined either by titrimetry (iodometrically) or by spectrophotometry by reacting it with a fixed concentration of tropaeolin 000 followed by the measurement of the dye colour at 490 nm. The amount of NBS reacted is related to the amount/concentration of DEC, which serves as basis of assyas. **Results:** The experimental variables affecting the assays were carefully studied and optimized. Titrimetry is applicable over 3-18 mg range and the reaction follows a 1:1 ratio (DEC:NBS). In spectrophotometry, Beer's law is obeyed over the concentration range of 15-120 µg mL^{-1} with a molar extinction coefficient of 1.65×10^3 L mol^{-1} cm^{-1}. The calculated limits detection (LOD) and quantification (LOQ) are found to be 0.44 and 1.33 µg mL^{-1}, respectively. Analysis of pure drug solution at three levels by the proposed methods had intra-day and inter-day coefficient of variation of <3% and the corresponding bias results were <2%. The methods were also validated for robustness, ruggedness and selectivity. **Conclusion:** The methods were successfully applied to the determination of DEC in tablets and syrup and the results compared well with the label claim and those obtained by the reference method. The study demonstrated the use of NBS and tropaeolin 000 as reagents in the titrimetric and spectrophotometric assay of DEC in pharmaceuticals.

Key words: Anti-parasitic agent, determination, bromination reaction, tropaeolin 000, formulations

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Diethylcarbamazine citrate (DEC), chemically called N,N-diethyl-4-methyl-1-piperazinecarboxamide citrate, is the drug of choice for treatment of lymphatic filariasis, a disease which affects people in the tropical areas of the world[1-3]. It is estimated that over 80 million people are infected worldwide[1]. Lymphatic filariasis results from infection with the parasitic roundworms *Wuchereria bancrofti*, *Brugia malayi* and *B. timori*. The infection is transmitted by mosquitoes. The adult parasites (microfilariae) live in the lymphatics, whereas, the larval stages (microfilariae) circulate in the blood-stream. Chronic manifestations of the disease include hydrocele, lymphedema and elephantiasis, which may affect more than 50% of the adult population in affected areas. Mass treatment with DEC is recommended for control of lymphatic filariasis[1] and in this connection, there is a need for an assay which could be used for the purpose of medication control.

Special attention has been paid to the assay of DEC in pharmaceuticals because of its extensive use in human and veterinary use. Several methods reported for the determination of DEC in body fluids and pharmaceuticals up to 2010 have been surveyed[4]. Recently, DEC in human plasma has been determined by liquid chromatography-mass spectrometry[5] for clinical and pharmacokinetic studies. The drug is official in British Pharmacopoeia[6], which describes a non-aqueous titration method for its determination. The United States Pharmacopoeia[7] uses a liquid chromatographic method with phosphate buffer-methanol system as the mobile phase for the assay with uv detection at 220 nm. A survey of the literature revealed that few methods have been reported for its determination in pharmaceuticals. High performance liquid chromatographic determination of DEC in combination with cetirizine[8-10], cetirizine and guaiphenesin[11] and chloropheniramine maleate[12,13] has been reported by several groups of researchers. Simultaneous determination of DEC and chlorpheniramine maleate in combined dosage forms has been accomplished by UV-spectrophotometry[14]. Although these methods are sensitive and selective, some of them are time consuming, complicated and require expensive instrumentation. Further, all the methods reported for pharmaceuticals[8-14] are applicable to combined dosage forms and are less suited for single-component dosage forms.

Titrimetry and spectrophotometry, because of their low cost, simplicity, speed and ease of performance, can be regarded as viable alternatives to the sophisticated instrument-oriented techniques. Recently, Weaver *et al.*[15] have described a low-tech analytical method for DEC in medicated salt, in which the medicated salt dosed with DEC is sprayed with a solution of potassium iodate and liberated iodine was determined by titrimetry. Very recently, one more iodometric titrimetric method using iodate-iodide mixture as the reagent has been reported by Swamy *et al.*[16]. Few visible spectrophotometric methods based on redox, ion-pair and charge-transfer complex reactions have been reported for DEC in single component dosage forms. The blue coloured chromogen formed due to the reduction of Folin-Ciocalteu (FC) reagent by DEC in alkaline medium was measured at 760 nm, which served as basis for the assay of DEC in pharmaceuticals, as reported by Swamy *et al.*[4]. The DEC is reported to form yellow-coloured ion-pair complexes with sulphonphthalein dyes, such as bromophenol blue and bromothymol blue[17] and bromocresol green and bromocresol purple[18] in chloroform and these reactions were successfully used to assay DEC, without involving extraction step. Two methods, based on formation of coloured charge-transfer complex[19] by DEC, with 2,3-dichloro-5,6-dicyano-p-benzoquinone (DDQ) or 2,4-dinitrophenol (DNP) in chloroform have also been described. The iodate-iodide mixture used as a titrimetric reagent[16] was extended to the spectrophotometric determination, in which the liberated iodine was either measured directly at 370 nm or complexed with starch and the iodine-starch complex measured at 570 nm.

Whereas, the titrimetric method of Weaver *et al.*[15] is not applicable for pharmaceuticals, the iodometric method[16] employs large quantities of saturated solutions of iodate and iodide, which are expensive and hence not suited for routine use. For the same reasons, two spectrophotometric methods[16] employing iodate-iodide mixture have limited applications. Methods based on ion-pair[17,18] and C-T complex[19] reactions are not green, since they require considerable amounts of organic solvents, which creates waste disposal problem. It is apparent that less cumbersome, simple and inexpensive methods are required for the determination of DEC in pharmaceuticals.

Thus, the present study is aimed at developing titrimetric and spectrophotometric methods for DEC that would overcome most of the limitations of the reported methods. The methods employ NBS as the bromination agent and are based on the determination of residual NBS by titrimetry and spectrophotometry, after allowing the drug to react with a known excess of NBS in HCl medium for a predetermined time. The methods were successfully applied to bulk sample and commercial dosage forms.

MATERIALS AND METHODS

Reagents: All reagents and chemicals used were of analytical grade and all solutions were freshly prepared daily in double distilled water. Pure diethylcarbamazine citrate, certified to be 99.86% pure, was procured from Inga Laboratories Pvt. Ltd., Mumbai, India and used as received. Banocide forte tablets (Glaxo Smith Kline Pharma. Ltd., Nashik, India) containing 50 and 100 mg DEC per tablet, Banocide syrup (Glaxo Smith Kline Pharma. Ltd., Bangalore, India) containing 120 mg DEC/5 mL and DECET tablets containing 150 mg DEC per tablet (from RND Laboratories, India) were purchased from local commercial sources. Approximately 0.01 M NBS solution was prepared by dissolving about 1.78 g of freshly crystallized chemical (Loba Chemie, Mumbai, India) in 1 L of water and standardized iodometrically[20] and diluted to 5 mM level with water and used in titrimetry. For spectrophotometric work, stock solution after standardization was diluted to 300 μg mL^{-1} concentration with water. Standard solution was stored in amber cloured bottle and kept in a refrigerator when not in use. About 2.5 g of sodium thiosulphate (S.D. Fine Chem., Mumbai, India) was dissolved in and diluted with water to one litre to get 0.01 M solution. Potassium iodide (10%) was prepared by dissolving 10 g of the chemical (Merck, Mumbai, India) in 100 mL water. Starch indicator (1%) was prepared by adding a paste of 1 g starch (Potato starch, Loba Chemie, Mumbai, India) in water to 100 mL boiling water, boiled for 1 min and cooled. Hydrochloric acid (5 and 3 M) was prepared by appropriate dilution of the concentrated acid (Merck, Mumbai, India, Sp. Gr. 1.18) with water. Tropaeolin 000, TRN (200 μg mL^{-1}) was prepared by dissolving accurately weighed 20 mg of dye (Loba Chemie, Mumbai, India) in water and diluted to 100 mL in a calibrated flask.

Standard drug solution: About 2 mg mL^{-1} stock standard solution was prepared by dissolving 500 mg of pure DEC in water and diluted to 250 mL in a calibrated flask and used in titrimetry. The stock solution was appropriately diluted stepwise with water to get a working concentration of 300 μg mL^{-1} for spectrophotometric assay.

Apparatus: A Systronics model 166 digital spectrophotometer (Systronics, Ahmedabad, Gujarat, India) with matched 1 cm quartz cells was used for absorbance measurements.

General procedures

Procedure for bulk drug

Titrimetry: About 10 mL aliquot of pure drug solution containing 3-18 mg DEC was measured accurately and transferred into a 100 mL glass-stoppered Erlenmeyer flask and acidified with 5 mL of 3 M HCl. Ten milliliters of 5 mM NBS solution was added to the flask by means of a pipette, the contents were mixed well and flask was set aside for 15 min with occasional swirling. Then, 5 mL of 10% KI solution was added and the liberated iodine was titrated with thiosulphate using starch indicator. A blank titration was performed and the mg of DEC in the measured aliquot was calculated from:

$$\text{DEC (mg)} = \frac{VMR}{n}$$

where, V is milliliter of NBS reacted, M is molarity of NBS, R is relative molecular mass of DEC and n is number of moles of NBS reacting with each mole of DEC.

Spectrophotometry: Different aliquots (0.5, 1.0,....., 4.0 mL) of 300 μg mL^{-1} standard drug solution were accurately measured into a set of 10 mL calibration flasks and the volume was brought to 4.0 mL with water. The solution was acidified by adding 1 mL of 5 M HCl and to each flask was added 1 mL of 300 μg mL^{-1} NBS. The contents were mixed and the flasks were set aside for 15 min with occasional shaking. Finally, 1 mL of 200 μg mL^{-1} tropaeolin 000 solution was added to each flak, volume was brought to mark with water, mixed and absorbance was measured at 490 nm vs. reagent blank after 5 min.

A standard graph was prepared by plotting absorbance vs concentration and the concentration of the unknown was computed using the regression equation derived from the Beer's law data.

Procedure for dosage forms

Tablets: Twenty tablets were weighed accurately and finely powdered. A portion of the powder equivalent to 100 mg of DEC was transferred into a 100 mL calibrated flask, 60 mL of water was added and the flask shaken for 15 min. Then, the volume was diluted to the mark with water, mixed well and the insoluble residue was filtered off using Whatman No. 42 filter paper. First 10 mL of the filtrate was discarded and 10 mL of the subsequent portion was analyzed in five replicates following the recommended titrimetric procedure. The tablet extract (1000 μg mL^{-1} in DEC) was diluted to 300 μg mL^{-1} with water and 2.0 mL aliquot was subjected to analysis (n = 5) following the spectrophotometric procedure.

Syrup: About 5 mL aliquot of the syrup containing 120 mg of DEC was accurately measured into a 100 mL calibrated flask, 60 mL of water added and shaken for 5 min before the volume

was diluted to the mark with water, mixed well and filtered using Whatman No. 42 filter paper. Subsequently, the steps described under procedure for tablets were followed.

Procedure for placebo and synthetic mixture: Inactive ingredients normally present in tablets, viz., starch (20 mg), talc (30 mg), calcium gluconate (20 mg), methyl cellulose (10 mg), lactose (10 mg), sodium alginate (10 mg), magnesium stearate (10 mg) and gelatin (10 mg) were mixed to get a homogeneous mixutre. Then, 100 mg of placebo was transferred to a 100 mL calibrated flask and its aqueous extract was prepared as described under "Procedure for tablets". Ten milliliters of the placebo extract was assayed by titrimetry and 2 mL of the diluted extract was analyzed by spectrophotometry as described earlier. About 100 mg of the placebo was added 100 mg of pure DEC to obtain of synthetic mixture and mixed well for uniform composition. The mixture was quantitatively transferred into a 100 mL calibrated flask and the steps described under "Procedure for tablets" were followed.

RESULTS AND DISCUSSION

The oxidizing as well as brominating ability of NBS has been widely for the titrimetric and spectrophotometric assay of several pharmaceutical substances[21-31]. However, its reaction with DEC has not been investigated yet. Therefore, the present study was devoted to apply NBS as a brominating agent for the development of titrimetric and spectrophotometric methods for the determination of DEC. The reaction between DEC and NBS in acid medium was found to be slow and hence, direct methods were not possible. The methods described are indirect and entail the determination of residual NBS after allowing the reaction between DEC and oxidant to go to completion under the specified experimental conditions. The amount of NBS reacted corresponds to the drug content in both methods. In titrimetry, the surplus oxidant was determined by iodometry and in spectrophotometry, it was determined by reacting with a fixed concentration of tropoaelin 000 dye. The method makes use of bleaching action of NBS on the dye, the decolouration being caused by the oxidative destruction of the dye. The DEC, when added in increasing concentrations to a fixed concentration of NBS, consumes the latter proportionately and there occurs a concomitant fall in the concentration of NBS. When a fixed concentration of dye is added to decreasing concentrations of NBS, a concomitant increase in the concentration of dye is output. Consequently, a proportional increase in the absorbance at the λ max is observed with increasing concentration of DEC (Fig. 1). Figure 2 shows the reaction pathways of assays. In spectrophotometry, preliminary experiments were performed to determine the concentration of TRN that would yield a reasonably high absorbance in acid medium and this was found to be 20 μg mL^{-1}. About 30 μg mL^{-1} NBS was found optimum to bleach the orange red colour due to 20 μg mL^{-1} tropaeolin in acid medium completely. Hence, different concentrations of DEC were reacted with 1 mL of 300 μg mL^{-1}

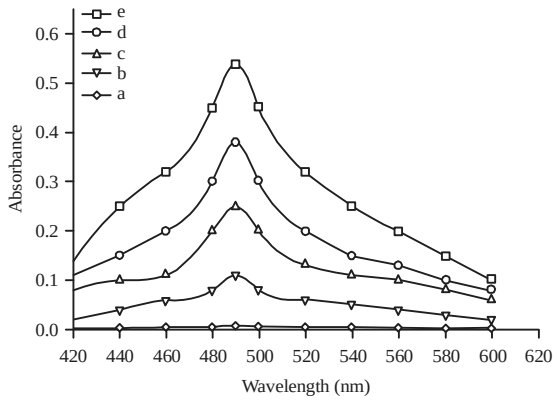

Fig. 1: Absorption spectra of 20 μg mL^{-1} tropeaolin 000 in the presence of, a: 0.0, b: 30, c: 60, d: 90 and e: 120 μg mL^{-1} DEC, the amounts of other reactants remained constant

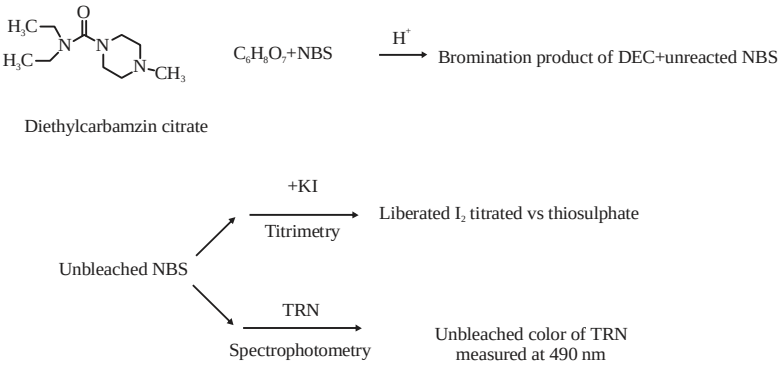

Fig. 2: Possible reaction pathways of titrimetric and spectrophotometric assay of DEC

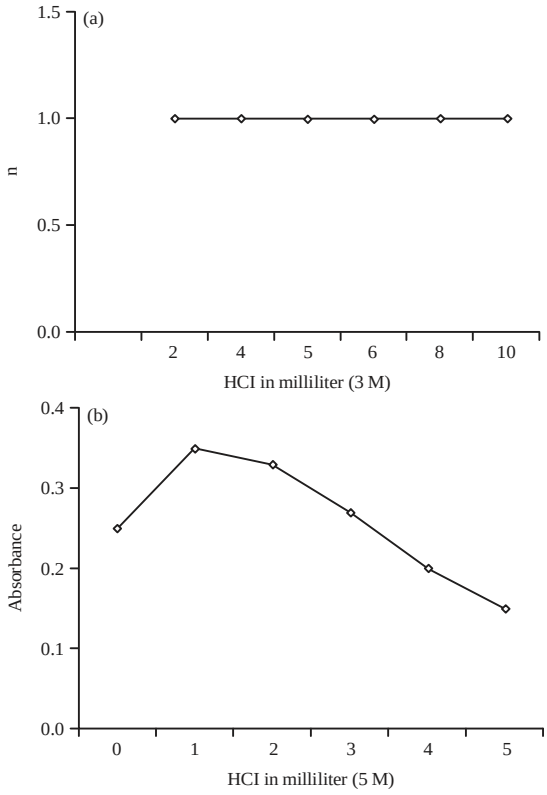

Fig. 3(a-b): Effect of HCl concentration with (a) 10 mg of DEC in titrimetry and (b) 75 µg mL^{-1} DEC in spectrophotometry (amounts of other reactants remained constant)

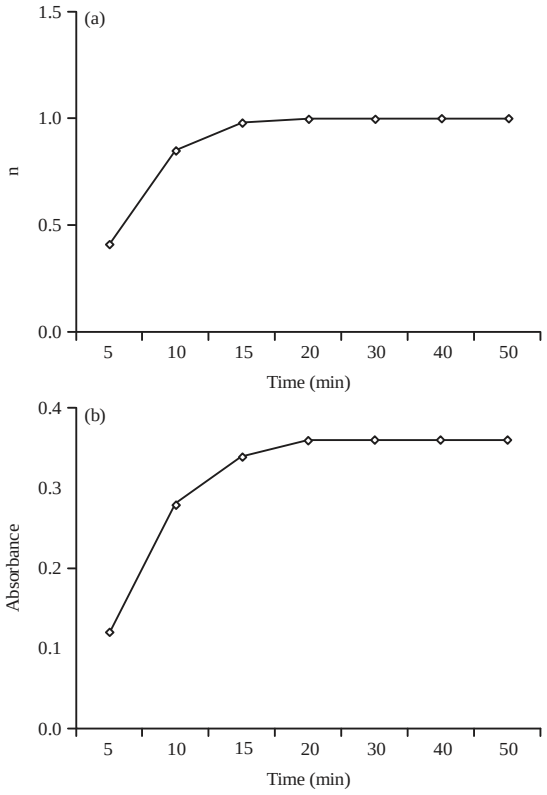

Fig. 4(a-b): Effect of reaction time with (a) 10 mg DEC (titrimetry) and (b) 75 µg mL^{-1} DEC (spectrophotometry)

NBS and the unreacted NBS was reacted with 1 mL of 200 µg mL^{-1} TRN in a total volume of 10 mL. This enabled the determination of DEC concentration range over which the method could be applied.

Method development: Experimental variables, such as, nature and strength of acid, reaction/standing time and concentrations of NBS and TRN dye, which affect the reaction between DEC and NBS and also determination of the residual NBS were investigated and optimized. These were studied by means of controlled experiments varying one parameter at a time.

Effect of acid medium: Of the several acids tried, HCl was found most suitable for the bromination of DEC and subsequent determination of residual NBS by iodometric titration and spectrophotometry using tropaeolin 000. Five milliliters of 3 M HCl in titrimetry (total 25 mL to begin with) and 1 mL of 5 M acid in spectrophotometry, in a 10 mL final volume, yielded best results (Fig. 3).

Effect of reaction time: A 15 min reaction time was found necessary for quantitative reaction between DEC and NBS in both methods and this was not critical, since reaction stoichiometry in titrimetry and the absorbance in spectrophotometry remained constant upto 30 min as shown in Fig. 4. In spectrophotometry, a standing time of 5 min was necessary for bleaching of the dye colour by residual NBS under the described optimum conditions.

Method validation

Linearity and sensitivity: Over the range investigated (3-18 mg in titrimetry), with 5 mM NBS a fixed reaction stoichiometry of 1:1 (DEC:NBS) was obtained, which served as the basis for calculations. In spectrophotometry, under optimum conditions, a linear relationship was found between absorbance and concentration in the range 15-120 µg mL^{-1} (Fig. 5). The calibration graph is described by the equation:

$$y = a+bx$$

where, y is absorbance, a is intercept, b is slope and x is concentration in µg mL^{-1} obtained by the method of least

squares. Correlation coefficient, intercept and slope for the calibration data are summarized in Table 1. Sensitivity parameters such as apparent molar absorptivity and sandell sensitivity and the limits of detection LOD and quantification LOQ calculated as per the current ICH guidelines[32] are also compiled in Table 1.

Precision and accuracy: Relative standard deviation percentage (RSD%) as an indicator precision and relative error percentage (RE%) as a measure of accuracy of the suggested methods were evaluated by replicate assays at three amount/concentration levels. The results of inter-day and intra-day precision and accuracy are presented in Table 2 and indicate good repeatability and reproducibility, besides reasonably high accuracy.

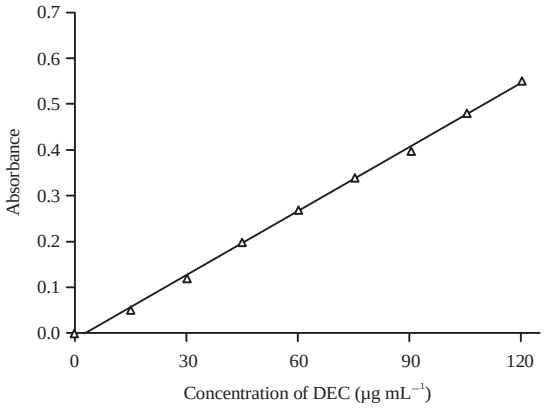

Fig. 5: Calibration graph

Table 1: Sensitivity and regression parameters (spectrophotometry)

Parameters	Values
λ_{max}, (nm)	490
Colour stability	30 min
Linear range (μg mL^{-1})	15-120
Molar absorptivity (ε) (L mol^{-1} cm^{-1})	1.65×10^3
Sandell sensitivity* (μg cm^{-2})	0.2376
Limit of detection (LOD) (μg mL^{-1})	0.44
Limit of quantification (LOQ) (μg mL^1)	1.33
Regression equation (y**)	
Intercept (a)	-0.0186
Slope (b)	0.0047
Standard deviation of a (S_a)	0.0998
Standard deviation of b (S_b)	0.00086
Regression coefficient (r)	0.9997

*Limit of determination as the weight in microgram per milliliter of solution, which corresponds to an absorbance of A = 0.001 measured in a cuvette of cross-sectional area 1 cm^2 and l = 1 cm, **y = a+bx, where, y is the absorbance, x concentration in microgram per milliliter a intercept and b slope

Robustness and ruggedness: For the evaluation of the method robustness, three optimized experimental variables, viz., acid volume and contact time (titrimetry) and contact time and wavelength (spectrophotometry), were slightly altered and their impact on the performance of the methods assessed. To study ruggedness, analysis was performed by three different analysts using the same procedure and also by a single analyst using three different burettes/cuvettes. The results of assays remained unaffected relative to those obtained under optimum conditions as shown by lower RSD% values (≤3%). These results are presented in Table 3.

Selectivity: To determine the selectivity of the described methods, placebo and synthetic mixture analyses were performed. When placebo blank was subjected to analysis by the proposed methods, no measurable NBS was consumed. When the synthetic mixture was subjected to analysis, at three levels by the proposed methods, the percent recoveries of pure drug ranged from 98.52±1.36 to 102.8±1.24 indicating non-interference from the inactive ingredients in the assays.

Application to dosage forms: The proposed methods were applied to the determination of DEC in two brands of tablets and one brand of syrup and the results are presented in Table 4. The same batch tablet powder and syrup were analyzed by the reference method (BP). The results were statistically compared by applying student's t and variance ratio F-test. The calculated student's t and F-values did not exceed the tabulated values at the 95% confidence level for four degrees of freedom, indicating agreeing accuracy and precision between the proposed methods and the reference method.

Table 2: Evaluation of intra-day and inter-day accuracy and precision

		Intra-day accuracy and precision (n = 7)			Inter-day accuracy and precision (n = 7)		
Method*	DEC taken	DEC found[a]	RSD[b] (%)	RE[c] (%)	DEC found	RSD[b] (%)	RE[c] (%)
Titrimetry	4	4.08	1.28	2.00	4.11	1.64	2.75
	8	8.12	1.33	1.50	8.09	1.55	1.13
	12	12.32	1.25	2.67	12.28	1.59	2.33
Spectrophotometry	30	30.67	1.59	2.23	30.52	1.84	1.73
	60	61.3	1.46	2.17	59.12	1.38	1.47
	90	91.5	1.37	1.67	91.22	1.46	1.36

[a]Mean value of 7 determinations, [b]Relative standard deviation (%), [c]Relative error (%), In titrimetry, DEC taken/found are in milligram and they are microgram per milliliter in spectrophotometry

Table 3: Method robustness and ruggedness expressed as intermediate precision (RSD%)

		Robustness (RSD%)			
		Parameters altered		Ruggedness (RSD%)	
Method	DEC taken*	**Acid/wavelength	***Contact time	Inter-burettes[#]/cuvettes[$] (RSD%) (n = 3)	Inter-analysis (RSD%) (n = 3)
Titrimetry	4	0.84	0.87	1.44	1.92
	8	0.74	1.65	1.56	1.72
	12	1.34	1.31	1.82	1.53
Spectrophotometry	30	1.76	0.88	1.49	1.92
	60	1.56	1.23	2.06	0.79
	90	1.43	0.97	1.75	1.82

*Titrimetry (mg) and spectrophotometry (μg mL^{-1}), **HCl volume used were: 4.5, 5.0 and 5.5 mL in titrimetry, wavelength used were: 485, 490 and 495 nm in spectrophotometry, ***Contact time used were: 14, 15 and 16 min in both methods, [#]In the case of titrimetry, [$]In the case of spectrophotometry

Table 4: Results of analysis of tablets by the proposed methods and statistical comparison of the results with the official method

		Found* (Percentage of nominal amount±SD)		
			Proposed methods	
Formulation analyzed	Nominal amount	Official method	Titrimetry	Spectrophotometry
Banocide forte tablets	50 mg per tablet	100.8±1.21	99.87±1.46	99.92.±0.76
			t = 1.10	t = 1.38
			F = 1.46	F = 2.53
Banocide forte tablets	100 mg per tablet	102.4±0.95	101.23±1.15	101.22.±0.86
			t = 1.76	t = 2.07
			F = 1.47	F = 1.22
DECET tablets	150 mg per tablet	101.2±1.34	99.83±1.25	100.42.±1.86
			t = 1.67	t = 0.76
			F = 1.15	F = 1.93
Banocide syrup	120 mg/5 mL	101.2 ±1.06	99.67±1.15	99.88±1.09
			t = 2.18	t = 1.93
			F = 1.18	F = 1.06

*Mean value of five determinations, tabulated t-value at the 95% confidence level and for four degrees of freedom is 2.77, tabulated F-value at the 95% confidence level and for four degrees of freedom is 6.39

Table 5: Results of recovery experiment through standard-addition method

Methods	Formulation studied	DEC in formulation	Pure DEC added	Total found	Pure DEC recovered percent±SD*
Titrimetry	Banocide forte tablets (100 mg)	6.07	3.0	8.99	99.82±1.34
		6.07	6.0	11.95	100.05±1.52
		6.07	9.0	14.88	99.22±1.44
	Banocide syrup	5.98	3.0	9.04	101.32±0.89
		5.98	6.0	11.95	100.60±1.62
		5.98	9.0	15.09	101.80±1.19
	Banocide forte tablets (100 mg)	40.49	20	61.20	102.13±1.54
		40.49	40	82.33	103.53±1.52
		40.49	60	103.25	103.62±1.64
Spectrophotometry	Banocide syrup	39.95	20	61.98	104.38±1.33
		39.95	40	81.89	102.85±0.97
		39.95	60	101.88	100.83±1.35

*Mean value of three determinations

Accuracy by recovery test: Pre-analyzed tablet powder and syrup were spiked with pure ETM at three levels and the total was determined by the proposed methods. The determination each level was replicated thrice. The results of percent recovery of drug which are an indication of accuracy are summarized in Table 5 and demonstrate that the methods are free from interference from the co-formulated substances. Comparison of present and existing methods with respect to performance characteristics is given in Table 6.

Table 6: Comparison of performance characteristics of the present method with the existing methods

Reagent used	Methodology	Linear range (μg mL^{-1})/ ε (L $mol^{-1}cm^{-1}$)	LOD/LOQ	Remarks	References
Folin-ciocalteu reagent	Blue coloured chromogen measured at 760 nm	10-100/2.08×10^3	1.06/3.2	-	Swamy *et al.*[4]
KIO_3-KI	I_2 measured at 370 nm	2.5-50/6.48×10^3	0.25/0.75	Saturated solution of	Swamy *et al.*[16]
KIO_3-KI/starch	I_2-starch measured at 570 nm	2.5-30/9.96×10^3	0.11/0.33	iodate-iodide required	
*BPB and BTB	Drug-dye ion-pair complex in chloroform measured at 430 and 440 nm	0.2-9.0/4.99×10^4	0.12/0.35	Use of organic solvent	Swamy *et al.*[17]
		0.4-14.0/2.32×10^3	0.08/0.24		
BCG and BCP	Drug-dye ion-pair complex in chloroform measured at 430 and 440 nm	0.4-12/2.11×10^4	0.25/0.76	Use of organic solvent	Swamy *et al.*[18]
		0.5-12.5/1.46×10^4	0.20/0.60		
DDQ and DNP	CT complex measured at 480 and 420 nm	4-90/1.88×10^3	0.21/0.63	Use of organic solvent	Swamy *et al.*[19]
		4-100/2.05×10^3	0.22/0.67		
NBS-Tropaeolin000	Measurement of residual dye colour at 490 nm	15-120/1.65×10^3	0.44/1.33	No use of organic solvent	Present study

*BPB: Bromophenol blue, BTB: Bromothymol blue, BCG: Bromocresol green, BCP: Bromocresol purple, DDQ: 2,3-dichloro-5,6-dicyano-p-benzoquinone, DNP: 2,4-dinitro phenol, CT: Charge transfer

CONCLUSION

The results demonstrate that micro level determination of diethylcarbamazine citrate is possible by titrimetry which can be performed with ease, rapidly and by using inexpensive chemicals. The method has the advantage of being applicable over a long range compared a narrow range offered by existing titrimetric method. Unlike most currently available spectrophotometric methods, the present method is free from unwelcome steps such as heating or extraction, pH adjustment and/or use of organic solvents. An additional advantage is that the absorbance measurement is made at longer wavelength (490 nm), where the interference from tablet excipients is expected to be less compared to shorter wavelengths (about 400 nm) used in most available methods. These advantages coupled with a fairly good accuracy and precision lend the methods aptly suitable for routine quality control.

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