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## SPECTROPHOTOMETRIC DETERMINATION OF REPAGLINIDE IN TABLETS BASED ON CHARGE-TRANSFER COMPLEXATION REACTION WITH CHLORANILIC ACID AND DICHLORO-DICYANO BENZOQUINONE

*Two simple, accurate, precise, inexpensive, selective and sensitive spectrophotometric methods are described for the determination of repaglinide (RPG) in bulk drug and its tablets. The methods were based on the charge-transfer complex reaction between RPG in acetonitrile with p-chloranilic acid (CAA) or 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) in dioxane and subsequent formation of intensely colored radical anions of the reagents which were measured at 520 (CAA) or 590 nm (DDQ). Several experimental variables affecting the complex formation, stability of the colored species and sensitivity of the reaction were optimized. Under the optimized conditions, Beer's law was obeyed over the concentration ranges of 20-400 and 5-80  $\mu\text{g ml}^{-1}$  RPG for CAA and DDQ methods, respectively, and the corresponding correlation coefficients were 0.9995 and 0.9998. The apparent molar absorptivity values were  $1.02 \times 10^3$  and  $4.60 \times 10^3$  for CAA and DDQ methods, respectively, with corresponding Sandell sensitivity values of 0.4438 and 0.0984  $\mu\text{g cm}^{-2}$ . Limits of detection (LOD) were calculated to be 7.07 and 2.42  $\mu\text{g ml}^{-1}$  and the limits of quantification (LOQ) were 21.43 and 7.33  $\mu\text{g ml}^{-1}$  RPG, for CAA and DDQ methods, respectively. Validation results demonstrated that the inter day and intra day accuracy were up to 97.56%. The precision determined did not exceed 2.5% of RSD. The methods were successfully used for the determination of RPG in tablet form and the results were in good agreement with the label claims as shown by the recoveries which were in the range of 99.22-102.8% with standard deviation values <2%. The accuracy of the methods was confirmed by recovery studies via standard-addition procedure with excellent recovery 98.24-104.0  $\pm$  1.08-3.35.*

*Keywords: repaglinide; p-chloranilic acid; 2,3-dichloro-5,6-dicyano-1,4-benzoquinone; charge transfer complexation; pharmaceuticals.*

Repaglinide (RPG), chemically known as (S)--(+)-2-ethoxy-4-[2-(3-methyl-1-[2-(piperidin-1-yl)phenyl]-butylamino)-2-oxoethyl] benzoic acid is a new carboxy methyl benzoic acid derivative. It is a novel prandial glucose regulator for the treatment type-2 diabetes mellitus [1,2]. It reduces the fasting glucose

concentration in patients with type-2 diabetes mellitus. It helps to control blood glucose sugar by increasing the amount of insulin released by the pancreas. RPG is rapidly absorbed from the gastrointestinal tract after oral administration. It differs from other antidiabetic agents in its structure, binding profile, duration of action and mode of extraction [3]. The drug is official in both European Pharmacopoeia [4], which describes non-aqueous titrimetry for its assay, and United State Pharmacopoeia [5] which recommends HPLC method for RPG determination. From the literature survey, it is revealed that a very few methods have been reported for the determination of RPG in

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pure form and dosage form; and include high performance liquid chromatography [6-8], reversed phase thin layer chromatography [9], spectrofluorimetry [8], voltammetry [10], UV-spectrophotometry [11-14] and visible spectrophotometry [14-16].

To the best of our knowledge, there are three reports on the use of visible spectrophotometry for the determination of RPG in pharmaceuticals [14-16], but all three reports are based on the formation of extractable ion association complex with different dyes. However, many of the above methods suffered from one or other disadvantage like poor sensitivity, measurements done at shorter wavelengths, extraction step, rigid pH control and use of expensive chemical and/or complicated experimental setup.

In this paper, the development and validation of two simple, sensitive, selective and rapid visible spectrophotometric methods are presented. The developed methods were based on the formation of intensely colored species resulting from the charge-transfer complex formed between RPG acting as an n-donor and CAA or DDQ acting as  $\pi$ -acceptor. These  $\pi$ -acceptors have numerous applications as analytical reagents and they have been used for the spectrophotometric determination of many organic compounds of pharmaceutical interest [17-21]. The developed methods were validated for linear range, LOD and LOQ, accuracy and precision and robustness and ruggedness. The validated methods were applied to commercially available tablets containing RPG with acceptable accuracy and precision.

## EXPERIMENTAL

### Apparatus

A Systronics model 106 digital spectrophotometer with 1 cm path length matched quartz cells was used for the absorbance measurement.

### Materials and reagents

Pure active ingredient sample of RPG was kindly supplied by Torrent Pharmaceuticals Ltd., Ahmadabad, India, as a gift. Eurepa-1 (1 mg RPG) and Eurepa-2 (2 mg RPG) tablets, marketed by Torrent Pharmaceuticals, Ahmadabad, India, were purchased from local commercial sources.

All chemicals used were of analytical reagent grade. Spectroscopic grade acetonitrile (Merck, Mumbai, India) was used to prepare RPG in both the methods while 1,4-dioxane (Merck, Mumbai, India) was used to prepare DDQ (Avra Synthesis Pvt. Ltd., Hyderabad, India) and CAA (Rolex Laboratory Reagent, Mumbai, India). A 0.2% (w/v) solution each of CAA

and DDQ was prepared freshly and separately by dissolving 0.2 g of each reagent in 100 ml of 1,4-dioxane.

### Standard RPG stock solution

A 500  $\mu\text{g ml}^{-1}$  RPG stock solution was prepared by dissolving 50 mg of pure drug in acetonitrile in a 100 ml volumetric flask and the solution was diluted to the mark with the same solvent and used for CAA method. The stock solution was diluted to get 100  $\mu\text{g ml}^{-1}$  working solution for use in method with DDQ.

### Procedures

#### CAA Method

Different aliquots (0.20-4.0 ml) of a standard 500  $\mu\text{g ml}^{-1}$  RPG solution were accurately transferred into a series of 5 ml calibrated flasks and the total volume in each flask was brought to 4 ml by adding acetonitrile. The content was mixed well after the addition of 1 ml of 0.2 % CAA solution and the absorbance was measured at 520 nm against a reagent blank similarly prepared without the addition of RPG.

#### DDQ Method

Into a series of 5 ml calibrated flasks, different aliquots (0.25-4.0 ml) of a standard 100  $\mu\text{g ml}^{-1}$  RPG solution were accurately transferred and the total volume were brought to 4 ml by adding adequate quantity of acetonitrile. To each flask, 1 ml of 0.2% DDQ solution was added and mixed well. After 2 min, the absorbance of the red colored C-T complex was measured at 590 nm against the reference blank similarly prepared.

#### Assay procedure for pharmaceutical formulation

Twenty five tablets were weighed and pulverized. An amount of tablet powder equivalent to 25 mg RPG was transferred into a 50 ml volumetric flask and about 30 ml of acetonitrile was added to the flask. The content was shaken well for 20 min and diluted to the mark with the same solvent. The resulting solution was filtered through Whatmann No. 42 filter paper and the filtrate was used for the assay by following the general procedure described under CAA method. The resulting tablet extract (500  $\mu\text{g ml}^{-1}$  in RPG) was diluted to 100  $\mu\text{g ml}^{-1}$  with acetonitrile and a suitable aliquot was used for the assay in DDQ method.

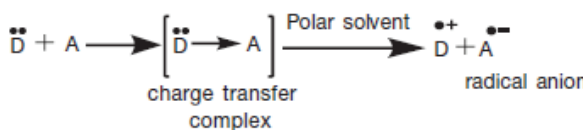
#### Procedure for the analysis of placebo blank and synthetic mixture

A placebo blank containing starch (10 mg), acacia (15 mg), hydroxyl cellulose (10 mg), sodium citrate (10 mg), talc (20 mg), magnesium stearate (15 mg) and sodium alginate (10 mg) was made and its solution was prepared as described under "Assay

procedure for pharmaceutical formulation” and then subjected to analysis. A synthetic mixture was separately prepared by adding pure RPG (25 mg) to the above mentioned placebo blank and the mixture was homogenized. The entire mixture containing 25 mg of RPG was taken and its extract was prepared as described above. Aliquots of extracts containing three different concentrations of RPG were subjected to assay by following the general procedures and the percentage recovery of RPG was evaluated.

## RESULTS AND DISCUSSION

The chemistry involved in the proposed methods was based on the reaction of RPG as n-electron donor with two  $\pi$ -acceptors, namely, CAA and DDQ to form charge transfer complexes which subsequently dissociate into radical anions depending on the polarity of the solvent used [22]. In polar solvents, such as acetonitrile, complete electron transfer from the donor to the acceptor takes place with the formation of intensely colored radical anions, according to the following equation:



In CAA method, the reaction of RPG as n-electron donor with CAA as a  $\pi$ -acceptor results in the formation of an intense orange-red product which exhibits absorption maxima at 520 nm in dioxane-acetonitrile medium (Figure 1). This can be attributed to the formation of the corresponding CAA radical anion

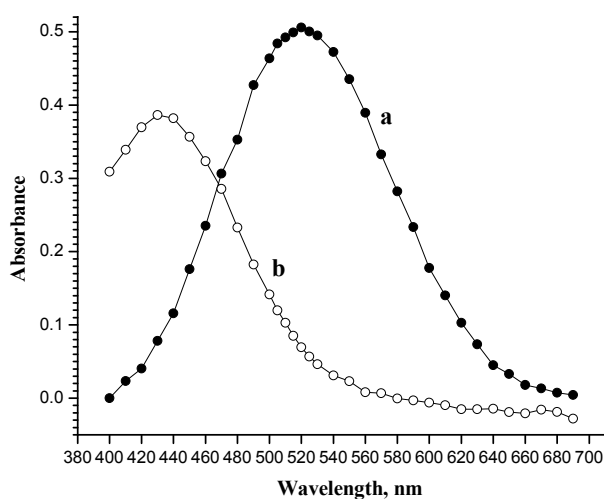


Figure 1. Absorption spectra of charge transfer complex of RPG-CAA ( $200 \mu\text{g ml}^{-1}$  RPG): a) RPG-CAA complex, b) CAA in 1,4-dioxane.

[23]. Similarly, in DDQ method, the interaction of RPG with DDQ in dioxane-acetonitrile at room temperature gave an intense reddish violet color which exhibit three maxima at 450, 550 and 590 nm due to the formation of the DDQ radical anions [24]. The wavelength 590 nm was selected for further studies because of higher sample absorbance and lower blank absorbance readings (Figure 2).

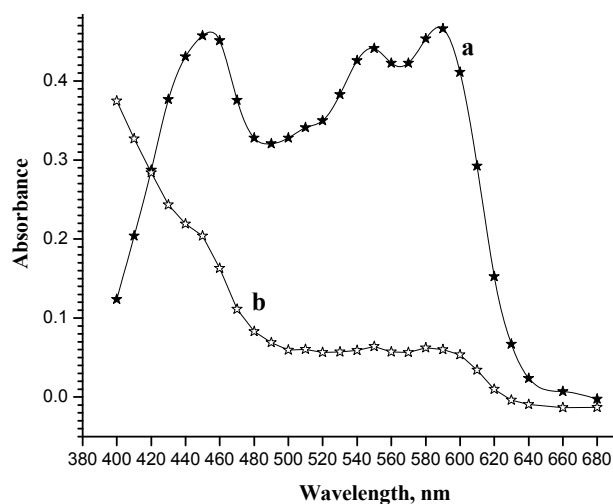


Figure 2. Absorption spectra of charge transfer complex of RPG-DDQ ( $50 \mu\text{g ml}^{-1}$  RPG): a) RPG-DDQ complex, b) DDQ in 1,4-dioxane.

### Optimization of reaction conditions

Optimum conditions were established by measuring the absorbance of C-T complexes at 520 and 590 nm, for CAA and DDQ methods, respectively, by varying one parameter at a time and keeping others constant. The concentrations of RPG used in the optimization studies are  $200 \mu\text{g ml}^{-1}$  RPG and  $50 \mu\text{g ml}^{-1}$  RPG in CAA and DDQ methods, respectively.

#### Effect of reagent concentration

To establish optimum concentrations of the reagents for the rapid formation of the stable charge-transfer complexes, RPG was allowed to react with different volumes of the reagents (0.5-3.0 ml of 0.2% CAA or 0.5-3.0 ml of 0.2% DDQ). In both the cases, maximum and minimum absorbance values were obtained for sample and blank, respectively, only when 1 ml of the reagent was used. At higher volumes of reagents the greater absorbance for blank and lower absorbance for C-T complexes were observed. Therefore, 1 ml of reagent in a total volume of 5 ml was used throughout the investigation.

#### Effect of solvent to dissolve drug and reagents

Acetonitrile was preferred to dissolve RPG than dichloromethane, acetone, 1,2-dichloroethane, 1,4-di-

oxane, methanol or ethanol because the complex formed in these solvents had very low absorbance values. Whereas in the case of reagents, highly intense colored products were formed when 1,4-dioxane medium was used as solvent to dissolve CAA and DDQ. Therefore, acetonitrile and 1,4-dioxane were chosen as solvents to dissolve RPG and the reagents, respectively.

#### *Effect of reaction time and stability of the C-T complexes*

The optimum reaction times were determined by measuring the absorbance of the formed complex upon the addition of reagent solution to RPG solution at room temperature. In both the methods the formation of C-T complexation reaction was instantaneous and the absorbance values of RPG-CAA and RPG-DDQ complexes were stable for at least 5 h and 75 min, respectively.

#### **Investigation of composition of C-T complexes**

The reaction stoichiometry between the drug and CAA or DDQ was determined spectrophotometrically by applying mole ratio method [25]. The experiments were performed by mixing a fixed volume of RPG (1.0 ml of  $1.54 \times 10^{-3}$  M RPG) with different volumes of each reagent separately (0.25–2.00 ml of  $1.54 \times 10^{-3}$  M CAA or DDQ) and maintaining the total volume at 5.0 ml with acetonitrile. The plots obtained by the mole ratio method confirm the formation of RPG-CAA and RPG-DDQ complexes in a mole ratio of 1:1, where a break point at 1.0 was obtained (Figure 3). Even though the drug has two nitrogen

atoms, only one nitrogen atom will be used in the formation of C-T complexes, *i.e.*, the nitrogen present in the piperidine ring, which is more basic than the nitrogen attached to the carbonyl group which is electron withdrawing group. Based on this, a possible reaction pathway for the formation of C-T complex is proposed and shown in Scheme 1.

#### **Method validation**

##### *Linearity, sensitivity, limits of detection and quantification*

A linear correlation was found between absorbance at  $\lambda_{\max}$  and concentration of RPG in the ranges given in Table 1. Regression analysis of the Beer's law data using the method of least squares was made to evaluate the slope ( $b$ ), intercept ( $a$ ) and correlation coefficient ( $r$ ) for each system and the values are presented in Table 1. The optical characteristics such as Beer's law limits, molar absorptivity and Sandell's sensitivity values for both the methods are also given in Table 1. The limits of detection ( $LOD$ ) and quantitation ( $LOQ$ ) were calculated according to ICH guidelines [26] using the formulae:

$$LOD = 3.3S/b \text{ and } LOQ = 10S/b$$

where  $b$  is the slope of the calibration plot and  $S$  is the standard deviation of " $n$ " reagent blank determinations under the same conditions as for the sample analysis in the absence of the analyte (RPG) ( $n = 5$  and 6 for CAA and DDQ methods, respectively). The calculated  $LOD$  and  $LOQ$  are also presented in Table 1.

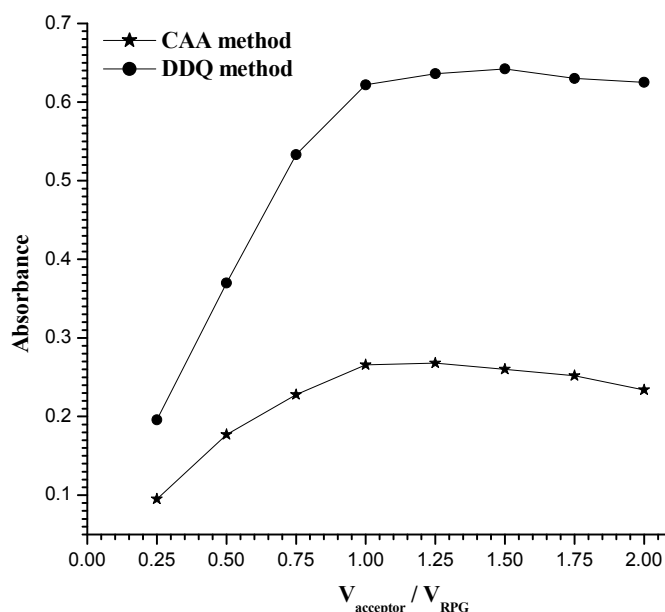
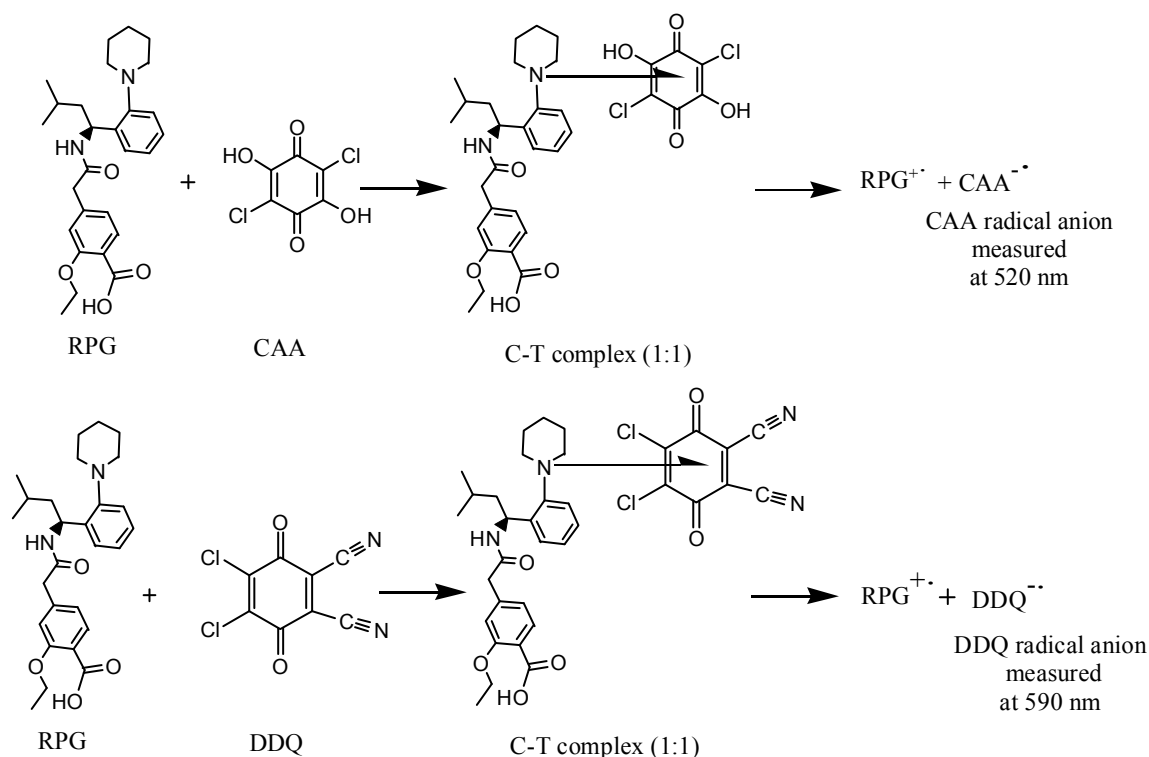


Figure 3. Molar ratio plot for RPG-CAA/DDQ ( $1.54 \times 10^{-3}$  M of RPG, CAA and DDQ) complexes.



Scheme 1. Possible reaction pathway for the formation of C-T complex between RPG and CAA/DDQ in acetonitrile-1,4-dioxane medium.

Table 1. Sensitivity and regression parameters

Parameter	CAA method	DDQ method
$\lambda_{\text{max}}$ / nm	520	590
Color stability	5 h	75 min
Linear range, $\mu\text{g ml}^{-1}$	20-400	5-80
Molar absorptivity ( $\epsilon$ ), $\text{L mol}^{-1} \text{cm}^{-1}$	$1.02 \times 10^3$	$4.60 \times 10^3$
Sandell sensitivity <sup>a</sup> , $\mu\text{g cm}^{-2}$	0.4438	0.0984
Limit of detection (LOD), $\mu\text{g ml}^{-1}$	7.07	2.42
Limit of quantification (LOQ), $\mu\text{g ml}^{-1}$	21.43	7.33
Regression equation, $Y^b$		
Intercept ( $a$ )	0.0046	-0.0132
Slope ( $b$ )	0.0022	0.0115
Correlation coefficient ( $r$ )	0.9995	0.9998
Standard deviation of $a$ ( $S_a$ )	$9.65 \times 10^{-3}$	$5.07 \times 10^{-3}$
Standard deviation of $b$ ( $S_b$ )	$4.00 \times 10^{-5}$	$1.10 \times 10^{-4}$

<sup>a</sup>Limit of determination as the weight in  $\mu\text{g}$  per ml of solution, which corresponds to an absorbance  $A = 0.001$  measured in a cuvette of cross-sectional area  $1 \text{ cm}^2$  and  $l = 1 \text{ cm}$ ; <sup>b</sup> $Y = a + bX$ , where  $Y$  is the absorbance,  $X$  is concentration in  $\mu\text{g/ml}$ ,  $a$  is intercept,  $b$  is slope

#### Precision and accuracy

The assays described under the construction of calibration curves were repeated seven times within the day to determine the intra-day precision and five times on different days to determine the inter-day precision of the methods. These assays were performed at three levels of analyte. The results of this study are summarized in Table 2. The percentage relative standard deviation (%RSD) values were

$\leq 1.51\%$  (intra-day) and  $\leq 2.34\%$  (inter-day) indicating high precision of the methods. Accuracy was evaluated as percentage relative error (RE) between the measured mean concentrations and taken concentrations for RPG. The percentage relative error was calculated at each concentration and these results are also presented in Table 2. Percent relative error (%RE) values of  $\leq 2.10\%$  demonstrates the high accuracy of the proposed methods.

Table 2. Evaluation of intra-day and inter-day accuracy and precision (%RE - percent relative error, %RSD - relative standard deviation,  $n$  = number of measurements)

Method	RPG taken, $\mu\text{g ml}^{-1}$	Intra-day accuracy and precision ( $n = 7$ )			Inter-day accuracy and precision ( $n = 5$ )		
		RPG found, $\mu\text{g ml}^{-1}$	%RE	%RSD	RPG found, $\mu\text{g ml}^{-1}$	%RE	%RSD
CAA	50	49.56	0.67	1.51	50.53	1.05	2.21
	100	100.11	1.52	0.76	100.6	1.42	1.61
	150	151.22	0.57	1.29	151.76	1.45	1.07
DDQ	20	20.23	2.10	0.90	20.25	1.14	1.02
	40	40.15	0.91	1.12	39.18	1.03	2.34
	60	60.54	1.21	1.50	59.59	0.15	1.54

### Selectivity

The results obtained from placebo blank and synthetic mixture analyses revealed that the inactive ingredients used in the preparation did not interfere in the assay of active ingredient. The absorbance values obtained from the placebo blank solution were almost equal to the absorbance of the blank which revealed no interference from the excipients. To study the role of additives added to the tablets, 2 ml each of the synthetic mixture solution (180 and 60  $\mu\text{g ml}^{-1}$  in RPG from CAA and DDQ methods, respectively) was assayed ( $n = 5$ ). The percentage recoveries of 96.7–103.6 with %RSD values in the range 0.86–2.11 demonstrated the accuracy as well as the precision of the proposed methods and complement the findings of the placebo blank analysis with respect to selectivity.

### Robustness and ruggedness

The robustness of the methods was evaluated by making small incremental changes in the volume of reagent, and the effect of the changes was studied on the absorbance of the complex systems. The changes had negligible influence on the results as revealed by small intermediate precision values expressed as %RSD  $\leq 0.69\%$ . Method ruggedness was demonstrated having the analysis done by four analysts, and also by a single analyst performing analysis on four different instruments in the same laboratory. Intermediate precision values (%RSD) in both instan-

ces were in the range 0.23–1.86 indicating acceptable ruggedness. The results are presented in Table 3.

### Application

The proposed methods were applied to the quantification of RPG in commercially available Eureka-2 (2 mg) and Eureka-1 (1 mg) tablets. The results obtained were compared with those obtained by a reference method [4] which involves the titration of the tablet extract in 1:6 (methanol:anhydrous acetic acid) with perchloric acid. Statistical analysis of the results did not detect any significant difference in the performance of the proposed methods and the reference method with respect to accuracy and precision as revealed by the Student's  $t$ -value and variance ratio  $F$ -value. The results of this study are given in Table 4.

### Recovery study

To further assess the accuracy of the proposed methods, recovery experiment was performed by applying the standard-addition technique. The recovery was assessed by determining the agreement between the measured standard concentration and added known concentration to the sample. The test was done by spiking the pre-analyzed tablet powder with pure RPG at three different levels (50, 100 and 150 % of the content present in the tablet powder (taken) and the total was found by the proposed methods. Each test was repeated three times. From this test the percentage recovery values were found in the range

Table 3. Method robustness and ruggedness expressed as intermediate precision (% RSD); the volumes of CAA or DDQ added were  $1.0 \pm 0.2$  ml

Method	RPG taken, $\mu\text{g ml}^{-1}$	Robustness		Ruggedness	
		Volume of the reagent, ml	Inter-analysts (%RSD) ( $n = 4$ )	Inter-instruments (%RSD) ( $n = 4$ )	
CAA	100	0.51	0.23	1.18	
	200	0.69	0.48	1.86	
	300	0.52	0.27	1.15	
DDQ	20	0.39	0.67	1.06	
	40	0.61	0.64	1.11	
	60	0.45	0.78	1.39	

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

Formulation name	Labeled amount <sup>a</sup>	Found <sup>b</sup> (Percent of label claim ± SD)		
		Reference method	CAA method	DDQ method
Eurepa 1	1	99.51±0.62	99.22±1.19	100.8±0.95
			$t = 0.48$	$t = 2.54$
			$F^d = 3.68$	$F = 2.35$
Eurepa 2	2	101.2±0.85	101.5±1.28	101.9±1.05
			$t = 0.44$	$t = 1.16$
			$F = 2.27$	$F = 1.53$

<sup>a</sup>Amount in mg per tablet; <sup>b</sup>mean value of 5 determinations; <sup>c</sup>tabulated  $t$ -value at the 95% confidence level and for four degrees of freedom is 2.77; <sup>d</sup>tabulated  $F$ -value at the 95% confidence level and for four degrees of freedom is 6.39

of 98.24-104.0% with standard deviation values from 1.08-3.35%. Closeness of the results to 100% showed the fairly good accuracy of the methods. These results are shown in Table 5.

The results are unaffected by slight variations in the optimized conditions such as reagent volume. Among the proposed methods, DDQ method was more sensitive than CAA method as can be seen from

Table 5. Results of recovery study via standard-addition method with tablets

Tablets studied	CAA method				DDQ method			
	RPG in tablet $\mu\text{g ml}^{-1}$	Pure RPG added $\mu\text{g ml}^{-1}$	Total found $\mu\text{g ml}^{-1}$	Pure RPG recovered <sup>a</sup> (percent ± SD)	RPG in tablet $\mu\text{g ml}^{-1}$	Pure RPG added $\mu\text{g ml}^{-1}$	Total found $\mu\text{g ml}^{-1}$	Pure RPG recovered <sup>a</sup> (percent ± SD)
Eurepa 1	99.22	50	150.2	102.0±2.57	20.16	10	30.46	103.0±3.12
	99.22	100	201.7	102.5±1.99	20.16	20	40.64	102.4±1.95
	99.22	150	254.5	103.5±1.08	20.16	30	51.09	103.1±1.57
Eurepa 2	101.5	50	150.6	98.24±2.20	20.38	10	30.78	104.0±3.35
	101.5	100	199.9	98.36±2.25	20.38	20	40.64	101.3±1.98
	101.5	150	250.8	99.56±1.45	20.38	30	51.25	102.9±1.84

<sup>a</sup>Mean value of three determinations

## CONCLUSIONS

Unlike the chromatographic and voltammetric methods, the proposed methods are simple and cost effective. The importance lies in the reactions upon which the procedures are based rather than the sophistication of the instrument. The proposed methods are superior to the existing UV-spectrophotometric methods in terms of selectivity as the interferences from many excipients present in the formulations will increase in the UV region compared with visible region. Also, all reported visible spectrophotometric methods are based on extractive spectrophotometric technique which has some difficulties and inaccuracies due to incomplete extraction or the formation of emulsions between the hydrocarbon solvent and the basic compound-containing solution. In addition, extractive spectrophotometric procedures are time consuming and required rigid pH control. The reagents used in the proposed methods are cheaper, easily available and the procedures do not involve any critical reaction conditions or tedious sample prepara-

tion. The results are unaffected by slight variations in the optimized conditions such as reagent volume. Among the proposed methods, DDQ method was more sensitive than CAA method as can be seen from

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NAUČNI RAD

## SPEKTROFOTOMETRIJSKO ODREĐIVANJE REPAGLINIDA U TABLETAMA ZASNOVANE NA REAKCIJI FORMIRANJA KOMPLEKSA UZ IZMENU NAELEKTRISANJA SA HLORANILNOM KISELINOM I DIHLORO-DICIJANO BENZOHINONA

*Razvijene su dve jednostavne, tačne, precizne, jeftine, selektivne i osetljive spektrofotometrijske metode za određivanje repaglinida (RPG) u rasutom stanju i tabletama. Metode su zasnovane na reakciji formiranja kompleksa uz izmenu naelektrisanja između RPG u acetonitrilu sa p-hloranilnom kiselinom (CAA) ili 2,3-dihloro-5,6-dicijano-1,4-benzokuinone (DDQ) u dioksanu pri čemu se grade intenzivno obojeni anjoni čiji se intenziteti mere na 520 (CAA) ili 590 nm (DDQ). Optimizovani su eksperimentalni uslovi koji utiču na formiranje kompleksa, stabilnost obojenih jedinjenja i osetljivost reakcije. Nadejno je da pod optimalnim uslovima za reakcije RPG sa CAA i DDQ važi Berov zakon u opsegu koncentracija 20-400 i 5-80  $\mu\text{g ml}^{-1}$ , odgovarajući koeficijenti korelacije su 0,9995 i 0,9998, molarne apsorbivnosti kompleksa su  $1,02 \times 10^3$  i  $4,60 \times 10^3 \text{ L mol}^{-1} \text{ cm}^{-1}$ , odgovarajući Sandelovi indeksi su 0,4438 i 0,0984  $\mu\text{g cm}^{-2}$ , limiti detekcije (LOD) su 7,07 i 2,42  $\mu\text{g ml}^{-1}$  i limiti kvantifikacije (LOQ) su 21,43 i 7,33  $\mu\text{g ml}^{-1}$ . Validacioni rezultati su pokazali da je tačnost određivanja u toku jednog i više dana iznad 97,56%. Preciznost određivanja ne prelazi 2,5% RSD. Metode su uspešno primenjene za određivanje RPG u obliku tableta, a rezultati su u dobroj saglasnosti sa vrednostima koje su propisane za lek što je pokazano i sa „recovery“ vrednostima koje su bile u rasponu od 99,22-102,8%, sa standardnom devijacijom vrednosti manjom od 2%. Tačnost metode je potvrđena metodom standardnog dodatka sa odličnim „recovery“ vrednostima koje su u opsegu 98,24-104,0 $\pm$ 1.08-3.35.*

*Ključne reči: repaglinid; p-hloranilna kiselina; 2,3-dihloro-5,6-dicijano-1,4-benzohinon; kompleksiranje uz izmenu naelektrisanja; farmaceutski preparati.*