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DEVELOPMENT AND VALIDATION OF UV VISIBLE SPECTROPHOTOMETER METHOD FOR ESTIMATION OF BEMPEDOIC ACID AND EZETIMIBE IN BULK DRUGS AND DOSAGE FORM

Nilesh V. Pawar[1]* and Sanjay Bhawar[2]

[1]Research Scholar, Bhagwant University, Ajmer, Rajasthan.

[2]Principal, Pravara College of Pharmacy, Loni, Ahmednagar, Maharashtra.

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***Corresponding Author**
Nilesh V. Pawar
Research Scholar, Bhagwant University, Ajmer, Rajasthan.

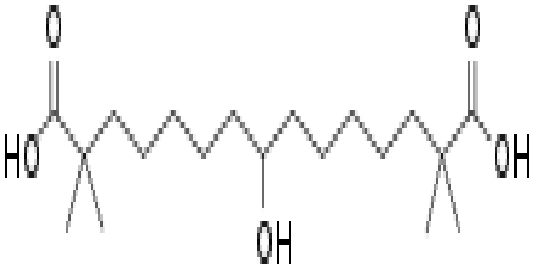
ABSTRACT

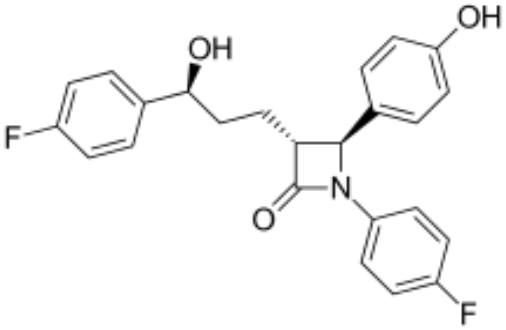
Aim: The objective of present investigation is to develop and validate UV visible spectrophotometer method for estimation of Bempedoic acid and Ezetimibe in marketed formulation. **Materials and Method:** Dissolution media was used for the development of UV visible spectrophotometer method. Both drugs Bempedoic acid and Ezetimibe was detected using 226nm wavelength. According to ICH requirements, the developed method was validated in terms of selectivity, linear range, precision, robustness, ruggedness, and reproducibility. The developed method was successfully used to estimate Bempedoic acid and ezetimibe in marked formulation. **Results:** Bempedoic acid and Ezetimibe has absorption wavelength of 226nm. Beer's law was followed at concentration ranging from 39.8995-299.2462 ppm for Bempedoic acid 2.2968 -17.226 ppm for Ezetimibe. The precision and repeatability scores were all within acceptable limits. Bempedoic acid and Ezetimibe recovery in marked formulation was found to be between 98-100%. The precision and repeatability values were within a 2% tolerance range. **Conclusion:** The method was found to be easy environmentally friendly, repeatable and cost effective and it can be used for Bempedoic acid and Ezetimibe analysis on regular basis.

KEYWORDS: Bempedoic acid, Ezetimibe, Dissolution media, UV visible spectrophotometer.

INTRODUCTION

The both drugs as Bempedoic acid and Ezetimibe are heterozygous familial and it is hypercholesterolemia. Bempedoic acid and Ezetimibe both are used together with diet to treat hypercholesterolemia and patients with heart disease who need extra lowering of their bad cholesterol levels. Bempedoic[1-2] acid is a strong inhibitor of ATP-citrate lyase, a target for LDL cholesterol lowering. Bempedoic acid is an alpha, omega-dicarboxylic acid that is pentadecane dioic acid which is substituted by methyl groups at position 2 and 14 and by a hydroxyl group at position 8. It is used to high LDL cholesterol. Ezetimibe[1-2] is an azetidine derivative, it prevents absorption of cholesterol by blocking the protein on epithelial cells of gastrointestinal tract, and in hepatocytes.





A) Structure of Bempedoic acid (Figure 1) **B) Structure of Ezetimibe (Figure 2)**

As a result, a UV visible spectrophotometer method for estimating Bempedoic acid and Ezetimibe in marketed formulation must be developed and validated.

MATERIALS AND METHOD

Instrumentation

For analytical method development of Bempedoic acid and Ezetimibe, UV-Spectrophotometer its was utilized. Table No.2

Table No.1: Instrumenst and Equipments used.

Sr. No.	Instruments	Make	Model/Series
1	Analytical Balance	Sartorius	CY224
2	pH-Meter	Mettler Toledo	LMPH-10
4	UV-Spectrophotometer	Lab India	550
5	Ultrasonicator	Bio Technics India	12L300H

Drug sample

Bempedoic acid and Ezetimibe was provided as a free sample by MSN lab Pvt Ltd in Hyderabad, and the marked formulation was obtained from the market.

Chemicals and Reagents

Table No. 2

Sr. No.	Name of chemical	Batch Number	Make	Expiry date/Use before
1.	Sodium acetate	J030A21	Rankem	July 2026
2.	Acetic acid	DE2P720623	Supelco	June 2025
3.	SLS	8122390124	Qualigens	March 2026
4.	Monosodium phosphate	M12M721315	Merck	April 2026
5.	Disodium phosphate	8175550224	Qualigens	April 2026

Selection of wavelength

Bempedoic acid and Ezetimibe was employed throughout the study since it is soluble in Dissolution media. Bempedoic acid 200ppm and Ezetimibe 11ppm both the working standard solution were scanned in the UV- Spectrophotometer between 200 nm to 400 nm, Q-absorption point was determined for both drugs. It is shown in 226 nm found as Q-absorption point.

Preparation of stock solution

Bempedoic acid: In order to prepare stock solution, weighed accurately 20 mg Bempedoic acid and transferred into 100 ml volumetric flask, added 15 ml of D.M and sonicated to dissolve the standard completely and diluted up to the mark with dissolution media (200 PPM).

Ezetimibe: In order to prepare stock solution, weighed accurately 11 mg of Ezetimibe and transferred into 100 ml volumetric flask, added 15 ml of solvent and sonicated to dissolve the standard completely and diluted up to the mark with dissolution media (116 PPM).
Further diluted 10 mL to 100 mL with dissolution media (11.6 PPM).

Method development

In Dissolution media, Bempedoic acid and Ezetimibe was shown to be soluble. As a result, this solvent was utilized to determine the detection wavelength and standard dealing concentration. The International Conference on Harmonization (ICH) has issued validation guidelines for analytical techniques, which characterize this method as characteristic performance verified through laboratory research. The developed technique was validated in accordance with ICH recommendations.

Specificity and Selectivity

Bempedoic acid and Ezetimibe has the Q-absorption at 226 nm, indicating that the procedure is selective and the spectra of the solvent revealed no absorbance at the wavelength of Bempedoic acid and Ezetimibe.

Linearity

Linearity was tested in the 39.8995-299.2462µg/ml range for Bempedoic acid and 2.2968-17.226µg/ml range for Ezetimibe. The standard stock solution prepared in Dissolution media and final dilution made with Dissolution media. Dilution of this solution were prepared respectively as per the linearity range.

Precision

To determine system precision, five absorbance of a solution containing 200µg/ml for Bempedoic acid and 11.11µg/ml for Ezetimibe were produced, with the absorbance of each solution measured at 226nm and the percentage Relative Standard Deviation (%RSD) was calculated. The precision of the method was determined by performing a sample dissolution under the conditions of the tests. Interday precision and intraday precision are two types of precision. Both precisions were calculated at stipulated time and % RSD were calculated.

Ruggedness

Ruggedness was determined by using a comparable planned procedure on a separate instrument and checking the reproducibility with different analysts.

Robustness

Robustness is done by doing the change in wavelength ±3nm, change in RPM ±5.

Accuracy

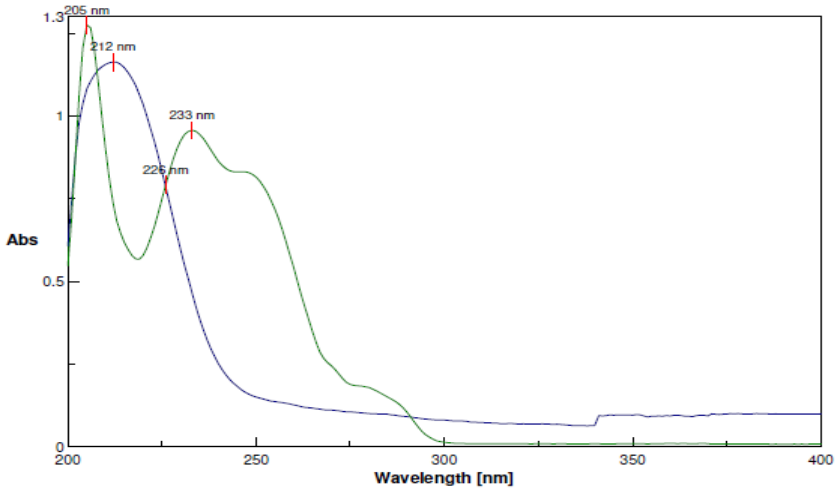
Accuracy was confirmed by doing recovery experiments in which the percent mean recovery of the sample was calculated using a standardization approach at three different levels: 50%, 100% and 150% of the sample solution. Dilutions were made from the above solution respectively. Three replicates of the concentration solution were prepared for each level, and a recovery study were conducted.

Analysis of marketed formulation:

Bempedoic acid and Ezetimibe in marketed formulation were determined using the established method.

Method development

Using UV-1800 and Dissolution media as a solvent, a UV-visible spectrometer technique was used for wavelength selection and shown in figure 3. The detail of the method established on UV were shown in Table 2.



Overlay UV spectrum of Bempedoic acid and Ezetemibe (Figure 3.)

Project Details:	Dissolution of Bempedoic acid	Dissolution of Ezetimibe
Label Claim	180 mg	10 mg
Dissolution media	0.05 M Phosphate buffer, pH 6.6	0.05M Sodium Acetate Buffer pH 4.5 with 0.45% SLS
Dissolution volume	900	900
Dissolution apparatus	Paddle	Paddle
Speed – RPM	50	50
Dissolution time (minutes)	45	45
Wavelength (nm)	226	226
Pathlength (mm)	1	1

Method validation

According to ICH requirements, the developed method was validated in terms of specificity, selectivity, linear range, precision, robustness, ruggedness and reproducibility.

Specificity and Selectivity

System suitability parameter is a very crucial part of all analytical methods. A standard solution of Bempedoic acid at a concentration of 200µg/ml and Ezetimibe at a concentration of 11µg/ml were taking absorbance five times. System suitability was tested by calculating the parameter like USP %RSD. The %RSD of the absorbance of Bempedoic acid and

Ezetimibe absorbance from five replicate were calculated. Bempedoic acid and Ezetimibe has the Q-absorption at 226nm, indicating that the technique is specific and selective (Figure 3).

Linearity

The calibration curve was obtained by plotting the response of Bempedoic acid and Ezetimibe against their respective concentration. Five different concentrations were prepared by serial dilution and used to obtained the calibration curve data ranging from 39.8995-299.2462µg/ml of Bempedoic acid and 2.2968-17.226µg/ml of Ezetimibe keeping the absorbance same throughout the study. The correlation coefficient ($R^{2)}$, slope and Y-intercept of Bempedoic acid and Ezetimibe were calculated.

Precision

System precision was evaluated in terms of inter-day precision (reproducibility) and intra-day (repeatability). The precision study was performed by absorbance six different sample in UV spectrophotometer on the same day and the next day. The value of standard deviation (SD) and %RSD were assessed from reproducibility and repeatability.

Accuracy/Recovery

The accuracy of an analytical method is the closeness of test results obtained by that method compared with the true values. Method accuracy was assessed by performing recovery studies for the pure drug by standard addition process. A known concentration of drug substance was added to three different concentration levels (50%, 100% and 150%). Control sample and recovery samples were absorbance in triplicate and % recovery was calculated at each level.

Limit of Detection (LOD) and Limit of Quantification (LOQ)

LOD and LOQ capability determine the sensitivity of the method. The calibration curve method was used to establish LOD and LOQ. Series of the dilute solution having known concentration were absorbance to determine the LOD and LOQ values. Signal to noise ratio was calculated for LOD and LOQ. %RSD of the absorbance was calculated at the QL concentration level.

$$LOD = \frac{3 \times \text{Standard deviation}}{\text{Slope of the calibration curve}} \qquad LOQ = \frac{10 \times \text{Standard deviation}}{\text{Slope of the calibration curve}}$$

Robustness

The measurement of the capacity of the analytical procedure to remain unaffected by small but deliberate variation in developed experimental conditions is called robustness. This parameter provides an indication that the developed method is robust and rugged. Different variables like dissolution RPM and wavelength were evaluated in the robustness study. Sample solution containing of Bempedoic acid and Ezetimibe substance and its drug product was an injected in actually condition as well as in all variable condition to check the robustness of the method. Effect on system suitability parameters like USP %RSD between Bempedoic acid and Ezetimibe and USP SD observed in the initial condition and each deliberate variation.

Solution stability

Solution stability was established at room temperature. These solutions were absorbance serially at respective time interval and the stability of these solution was evaluated for 24 hr. The comparison of these solutions was evaluated against the freshly prepared sample solution.

RESULTS AND DISCUSSION

Method validation

System suitability

The % RSD of the absorbance of Bempedoic acid and Ezetimibe absorbance in six replicate of system suitability solution was 0.00 and 0.00 (<2.0). The method was suitable for use as all the parameters were within the limit. The outcome of system suitability parameters was summarized in table 3 and Figure 3.

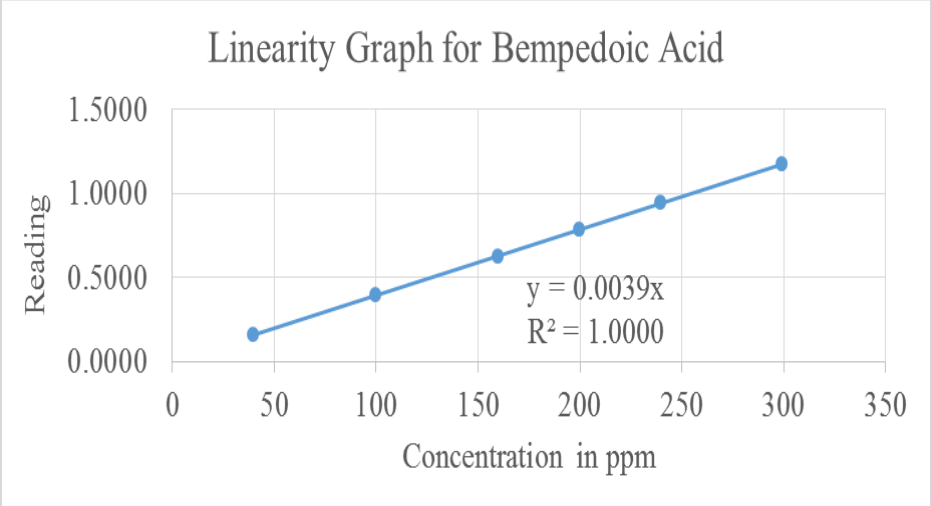
Sr. No.	System precision	Bempedoic Acid	Ezetimibe
		Absorbance	Absorbance
1	Standard preparation-Reading 1	0.7834	0.6731
2	Standard preparation-Reading 2	0.7834	0.6732
3	Standard preparation-Reading 3	0.7833	0.6730
4	Standard preparation-Reading 4	0.7834	0.6731
5	Standard preparation-Reading 5	0.7834	0.6731
	Mean	0.7833	0.6731
	SD	0.0000	0.0000
	%RSD	0.0000	0.0000

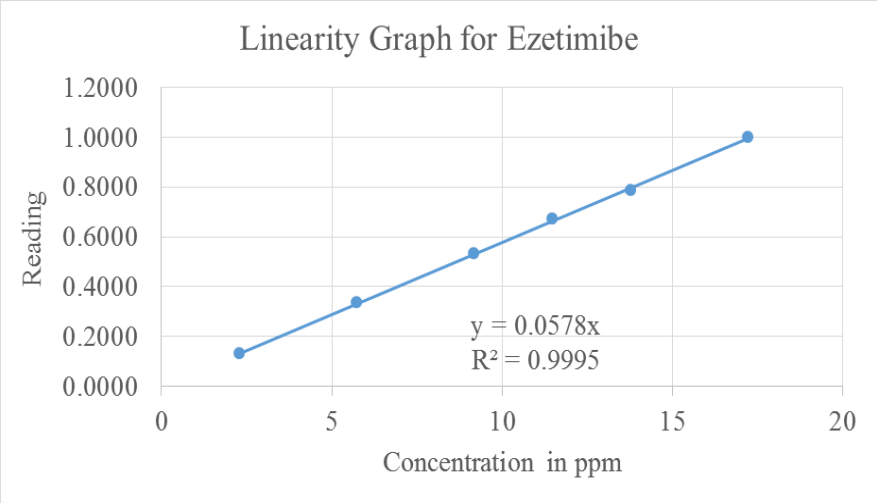
Linearity

Linearity of the method was performed by determining standard solution at six different concentration levels covering the range of 39.8995 to 299.2462 µg/ml for Bempedoic acid and 2.2968 to 17.226 µg/ml for Ezetimibe. Slope, coefficient of correlation (r^2) and intercept for Bempedoic acid were 0.0039, 0.9999 and 0.0012 and for Ezetimibe 0.0576, 0.9997 and 0.0025 respectively. Linearity data are shown in Table 5. The calibration curve of Bempedoic acid and Ezetimibe is shown in Figure5 and Figure 6.

Table 4

Bempedoic acid		**Ezetimibe**	
Concentration (µg/ml)	Absorbance	Concentration (µg/ml)	Absorbance
39.8995	0.1553	2.2968	0.1331
99.7487	0.3944	5.742	0.3357
159.598	0.6236	9.1872	0.5313
199.4975	0.7840	11.484	0.6732
239.397	0.9398	13.7808	0.7853
299.2462	1.1703	17.226	0.9990
Slope	0.0039		0.0576
Intercept	0.0012		0.0025
Correlation (r^2)	0.9999		0.9997





Precision: The % RSD values were 0.3909 and 0.3873 for Bempedoic acid and Ezetimibe respectively for precision and 0.3674 and 0.2255 for Bempedoic acid and Ezetimibe respectively for Intermediate precision and. Results of Intra and inter day precision of Bempedoic acid and Ezetimibe are presented in Table 6.

Table No. 5: For bempedoic acid.

Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.7655	99.6698
Sample 2	0.7589	98.8105
Sample 3	0.7643	99.5186
Sample 4	0.7621	99.2272
Sample 5	0.7676	99.9433
Sample 6	0.7641	99.4876
Mean		**99.4428**
STD DEV		**0.3888**
% RSD		**0.3909**
Intermediate Precision		
Sample	**Area**	**% Assay**
Sample 1	0.7542	97.5949
Sample 2	0.7597	98.3067
Sample 3	0.7551	97.7114
Sample 4	0.7585	98.1514
Sample 5	0.7611	98.4878
Sample 6	0.7598	98.3196
Mean		**98.0953**
STD DEV		**0.3605**
% RSD		**0.3674**
Precision plus intermediate precision	**Mean**	**98.7690**
	STD DEV	**0.7893**
	% RSD	**0.7991**

Table No. 6: For ezetimibe.		
Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.6543	100.4692
Sample 2	0.6520	100.1160
Sample 3	0.6591	101.2062
Sample 4	0.6578	101.0066
Sample 5	0.6549	100.5613
Sample 6	0.6555	100.6534
Mean		**100.6687**
STD DEV		**0.3899**
% RSD		**0.3873**
Intermediate Precision		
Sample	**Area**	**% Assay**
Sample 1	0.6465	100.8464
Sample 2	0.6441	100.4720
Sample 3	0.6467	100.8776
Sample 4	0.6472	100.9556
Sample 5	0.6437	100.4096
Sample 6	0.6461	100.7840
Mean		**100.7242**
STD DEV		**0.2272**
% RSD		**0.2255**
Precision plus intermediate precision	**Mean**	**100.6964**
	STD DEV	**0.3056**
	% RSD	**0.3034**

Accuracy/Recovery

The % recovery was obtained to be in the range of 20% to 150%. The recovery results showed that the developed method is precise and also observed that there was no intervention due to the existence of DPs. The results are shown in table 7.

Table No. 7

Accuracy-Bempedoic acid	Amount of standard added (mg)	Final Volume	Amount of standard added (mg) after purity correction	Reading	Amount found (mg)	% Recovery	Average	SD	%RSD
Level-1-20%-Sample-1	36.5120	900	36.3294	0.1532	35.9629	98.9911	99.6105	0.7922	0.7952
Level-1-20%-Sample-2	36.1270	900	35.9463	0.1539	36.1272	100.5032			
Level-1-20%-	36.4560	900	36.2737	0.1535	36.0333	99.3372			

Sample-3									
Level-2-50%-Sample-1	90.5120	900	90.0594	0.3822	89.7195	99.6225	100.1132	0.4618	0.4612
Level-2-50%-Sample-2	90.4580	900	90.0057	0.3841	90.1655	100.1775			
Level-2-50%-Sample-3	90.6720	900	90.2186	0.3864	90.7054	100.5395			
Level-3-100%-Sample-1	180.5300	900	179.6273	0.7612	178.6878	99.4769	99.6598	0.1603	0.1608
Level-3-100%-Sample-2	180.8120	900	179.9079	0.7643	179.4155	99.7263			
Level-3-100%-Sample-3	180.1540	900	179.2532	0.7619	178.8521	99.7762			
Level-4-150%-Sample-1	270.6510	900	269.2977	1.1423	268.1491	99.5734	99.8608	0.2618	0.2621
Level-4-150%-Sample-2	270.4590	900	269.1067	1.1455	268.9003	99.9233			
Level-4-150%-Sample-3	270.4210	900	269.0688	1.1472	269.2994	100.0857			

Table No. 8

Accuracy-Ezetimibe	Amount of standard added (mg)	Final Volume	Amount of standard added (mg) after purity correction	Reading	Amount found (mg)	% Recovery	Average	SD	%RSD
Level-1-20%-Sample-1	2.0221	900	2.0018	0.1321	2.0293	101.3737	100.0084	1.7727	1.7725
Level-1-20%-Sample-2	2.0151	900	1.9949	0.1307	2.0078	100.6466			
Level-1-20%-Sample-3	2.071	900	2.0502	0.1308	2.0093	98.005			
Level-2-50%-Sample-1	5.1273	900	5.076	0.3289	5.0525	99.537	100.241	0.6104	0.6089
Level-2-50%-Sample-2	5.2231	900	5.1708	0.3387	5.2031	100.6246			
Level-2-50%-Sample-3	5.1107	900	5.0595	0.3312	5.0879	100.5613			
Level-3-100%-Sample-1	10.1354	900	10.034	0.6634	10.1911	101.5656	101.6771	0.1747	0.1718
Level-3-100%-Sample-2	10.1409	900	10.0394	0.6658	10.228	101.8785			
Level-3-100%-Sample-3	10.1501	900	10.0485	0.6645	10.208	101.5873			
Level-4-150%-Sample-1	15.0956	900	14.9446	0.9875	15.17	101.5082	101.2835	0.6608	0.6524
Level-4-150%-	15.3475	900	15.194	0.9944	15.276	100.5396			

Sample-2									
Level-4-150%-Sample-3	15.2059	900	15.0538	0.9976	15.3252	101.8028			

Robustness

A robustness study was performed by creating small, deliberate variations in the speed (RPM) (±5), and change in initial wavelength (±3nm) and found to be unaffected upon these deliberate variations in parameters. Results are shown in Table 9 and Table 10.

Table No. 9: For bempedoic acid.		
Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.7655	99.6698
Sample 2	0.7589	98.8105
Sample 3	0.7643	99.5186
Sample 4	0.7621	99.2272
Sample 5	0.7676	99.9433
Sample 6	0.7641	99.4876
Mean		**99.4428**
STD DEV		**0.3888**
% RSD		**0.3909**
Robustness Low Wavelength (223nm)		
Sample	**Area**	**% Assay**
Sample 1	0.7541	98.2711
Sample 2	0.7538	98.2321
Sample 3	0.7597	99.0009
Sample 4	0.7566	98.5969
Sample 5	0.7530	98.1278
Sample 6	0.7590	98.9097
Mean		**98.5230**
STD DEV		**0.3708**
% RSD		**0.3763**
Precision plus Robustness Low wavelength (223nm)	**Mean**	**98.9829**
	STD DEV	**0.6016**
	% RSD	**0.6077**

Table No. 10: For bempedoic acid.		
Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.7655	99.6698
Sample 2	0.7589	98.8105
Sample 3	0.7643	99.5186
Sample 4	0.7621	99.2272
Sample 5	0.7676	99.9433
Sample 6	0.7641	99.4876

Mean		**99.4428**
STD DEV		**0.3888**
% RSD		**0.3909**
Robustness High Wavelength (229nm)		
Sample	**Area**	**% Assay**
Sample 1	0.7349	99.4201
Sample 2	0.7302	98.7843
Sample 3	0.7377	99.7989
Sample 4	0.7367	99.6636
Sample 5	07345	99.3660
Sample 6	0.7301	98.7707
Mean		**99.3006**
STD DEV		**0.4349**
% RSD		**0.4379**
Precision plus Robustness High wavelength (229nm)	**Mean**	**99.3717**
	STD DEV	**0.4003**
	% RSD	**0.4028**

Table No. 11: For ezetimibe.

Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.6543	100.4692
Sample 2	0.6520	100.1160
Sample 3	0.6591	101.2062
Sample 4	0.6578	101.0066
Sample 5	0.6549	100.5613
Sample 6	0.6555	100.6534
Mean		**100.6687**
STD DEV		**0.3899**
% RSD		**0.3873**
Robustness Low Wavelength (223nm)		
Sample	**Area**	**% Assay**
Sample 1	0.6427	100.3437
Sample 2	0.6438	100.5155
Sample 3	0.6402	99.9534
Sample 4	0.6451	100.7184
Sample 5	0.6430	100.3906
Sample 6	0.6419	100.2188
Mean		**100.3567**
STD DEV		**0.2603**
% RSD		**0.2593**
Precision plus Robustness Low wavelength (223nm)	**Mean**	**100.5127**
	STD DEV	**0.3556**
	% RSD	**0.3537**

Table No. 12: For ezetimibe.		
Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.6543	100.4692
Sample 2	0.6520	100.1160
Sample 3	0.6591	101.2062
Sample 4	0.6578	101.0066
Sample 5	0.6549	100.5613
Sample 6	0.6555	100.6534
Mean		**100.6687**
STD DEV		**0.3899**
% RSD		**0.3873**
Robustness High Wavelength (229nm)		
Sample	**Area**	**% Assay**
Sample 1	0.6701	101.1524
Sample 2	0.6689	100.9713
Sample 3	0.6605	99.7033
Sample 4	0.6639	100.2165
Sample 5	0.6610	99.7788
Sample 6	0.6672	100.7147
Mean		**100.4228**
STD DEV		**0.6153**
% RSD		**0.6127**
Precision plus Robustness High wavelength (229nm)	**Mean**	**100.5458**
	STD DEV	**0.5076**
	% RSD	**0.5048**

Table No. 13: For bempedoic acid.		
Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.7655	99.6698
Sample 2	0.7589	98.8105
Sample 3	0.7643	99.5186
Sample 4	0.7621	99.2272
Sample 5	0.7676	99.9433
Sample 6	0.7641	99.4876
Mean		**99.4428**
STD DEV		**0.3888**
% RSD		**0.3909**
Robustness Low RPM (45)		
Sample	**Area**	**% Assay**
Sample 1	0.7654	99.7434
Sample 2	0.7698	100.3171
Sample 3	0.7612	99.1964
Sample 4	0.7600	99.0400

Sample 5	0.7567	98.6100
Sample 6	0.7642	99.5873
Mean		**99.4157**
STD DEV		**0.5979**
% RSD		**0.6014**
Precision plus Robustness Low RPM (45)	**Mean**	**99.4292**
	STD DEV	**0.4811**
	% RSD	**0.4838**

Table No. 14: For bempedoic acid.

Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.7655	99.6698
Sample 2	0.7589	98.8105
Sample 3	0.7643	99.5186
Sample 4	0.7621	99.2272
Sample 5	0.7676	99.9433
Sample 6	0.7641	99.4876
Mean		**99.4428**
STD DEV		**0.3888**
% RSD		**0.3909**
Robustness High RPM (55)		
Sample	**Area**	**% Assay**
Sample 1	0.7715	100.6402
Sample 2	0.7713	100.6142
Sample 3	0.7712	100.6011
Sample 4	0.7789	101.6056
Sample 5	0.7767	101.3186
Sample 6	0.7757	101.1881
Mean		**100.9946**
STD DEV		**0.4337**
% RSD		**0.4294**
Precision plus Robustness High RPM (55)	**Mean**	**100.2187**
	STD DEV	**0.9005**
	% RSD	**0.8985**

Table No. 15: For ezetimibe.

Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.6543	100.4692
Sample 2	0.6520	100.1160
Sample 3	0.6591	101.2062
Sample 4	0.6578	101.0066
Sample 5	0.6549	100.5613
Sample 6	0.6555	100.6534
Mean		**100.6687**
STD DEV		**0.3899**
% RSD		**0.3873**

Robustness Low RPM (45)		
Sample	**Area**	**% Assay**
Sample 1	0.6516	101.7790
Sample 2	0.6498	101.4979
Sample 3	0.6471	101.0761
Sample 4	0.6420	100.2795
Sample 5	0.6474	101.1230
Sample 6	0.6479	101.2011
Mean		**101.1594**
STD DEV		**0.5065**
% RSD		**0.5006**
Precision plus Robustness Low RPM (45)	**Mean**	**100.9141**
	STD DEV	**0.5013**
	% RSD	**0.4967**

Table No. 16: For ezetimibe.		
Precision		
Sample	**Absorbance**	**% Release**
Sample 1	0.6543	100.4692
Sample 2	0.6520	100.1160
Sample 3	0.6591	101.2062
Sample 4	0.6578	101.0066
Sample 5	0.6549	100.5613
Sample 6	0.6555	100.6534
Mean		**100.6687**
STD DEV		**0.3899**
% RSD		**0.3873**
Robustness High RPM (55)		
Sample	**Area**	**% Assay**
Sample 1	0.6600	101.3743
Sample 2	0.6673	102.4955
Sample 3	0.6617	101.6354
Sample 4	0.6613	101.5740
Sample 5	0.6682	102.6338
Sample 6	0.6643	102.0347
Mean		**101.9579**
STD DEV		**0.5184**
% RSD		**0.5084**
Precision plus Robustness High RPM (55)	**Mean**	**101.3133**
	STD DEV	**0.8028**
	% RSD	**0.7923**

LOD and LOQ: Detection Limit (LOD) and Quantitation Limit (LOQ)

Remark: This parameter is not applicable.

Solution Stability and

% RSD of Bempedoic acid and Ezetimibe were found less than 2.0 % and there were no major changes observed in absorbance indicates solution stability up to 48 hr. Both drugs were shown in Table No.17 and 18.

Table No.17: Solution stability of bempedoic acid.				
% Absolute difference	Initial	12 hrs.	24 hrs.	48 hrs.
Standard solution	NA	0.0127	0.0255	0.0255
Sample solution	NA	0.3528	0.4965	0.6533

Table No. 18: Solution stability of ezetimibe.				
% Absolute difference	**Initial**	**12 hrs.**	**24 hrs.**	**48 hrs.**
Standard solution	NA	0.1038	0.2076	0.4598
Sample solution	NA	0.3832	0.6746	0.9658

CONCLUSION

A novel, linear, accurate, specific, reliable and specific UV Spectrophotometer method was developed and fully validated for the determination of Bempedoic acid and Ezetimibe in the presence of its drug products. Validation of the UV Spectrophotometer method as per the ICH guidelines demonstrates that the method is highly sensitive, linear, rapid and robust. Therefore, work may be useful for estimation of Bempedoic acid and Ezetimibe in bulk and pharmaceutical dosage form.

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ABBREVIATIONS

°C: Degree Celsius; **RPM**; revolution per minutes **µg:** Microgram; **ml:** Millilitre; **hr:** Hour; **Abs;** Absorbance **UV:** Ultra Violet; **min:** Minutes; **NA:** Not Applicable.

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