

# New LC–UV Methods for Pharmaceutical Analysis of Novel Anti-diabetic Combinations

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Received: 01 August 2016; accepted: 24 September 2016

New, simple, selective, and sensitive liquid chromatography–ultraviolet (LC–UV) methods have been developed and subsequently validated for simultaneous determination of linagliptin–empagliflozin combination and simultaneous determination of alogliptin benzoate–pioglitazone hydrochloride combination. Linearity was found to be acceptable over the concentration ranges of 2–50  $\mu\text{g mL}^{-1}$ , 4–100  $\mu\text{g mL}^{-1}$ , 0.5–25  $\mu\text{g mL}^{-1}$ , and 1–25  $\mu\text{g mL}^{-1}$  for linagliptin (LNG), empagliflozin (EMG), alogliptin (ALG), and pioglitazone (PGN), respectively. All the methods were applied successfully to the analysis of the pharmaceutical dosage forms. The optimized methods were validated and proved to be robust and accurate for the quality control of the mentioned drugs in their different pharmaceutical dosage forms.

**Keywords:** linagliptin, empagliflozin, alogliptin benzoate, pioglitazone hydrochloride, LC–UV

## 1. Introduction

Empagliflozin (EMG) (Figure 1a) is an inhibitor of sodium glucose co-transporter-2 (SGLT-2), so it inhibits glucose re-absorption into the blood [1]. Linagliptin (LNG) (Figure 1b) and alogliptin (ALG) (Figure 1c) are inhibitors to dipeptidyl peptidase-4 (DPP-4), so they stimulate insulin release [2]. Pioglitazone (PGN) (Figure 1d) is one of the thiazolidinediones which increase insulin sensitivity [3].

To the best of the authors' knowledge, one chromatographic method [4] and one spectrophotometric Vierodt's method [5] were described for simultaneous determination of EMG and LNG. Furthermore, simultaneous equation and partial least square 2 (PLS-2) methods were used for simultaneous determination of EMG and metformin [6]. Moreover, the literature showed few chromatographic methods [7–10] and one spectrophotometric method for simultaneous determination of ALG and PGN based on first derivative and dual wavelength techniques [11].

Two new proposed methods have been discussed in the present work. The aim of the developed high-performance liquid chromatography (HPLC) methods is to present a new simple HPLC method for simultaneous determination of LNG and EMG in bulk powder, in their different ratios as laboratory-prepared mixtures and in their pharmaceutical dosage form (HPLC-1), and to present another accurate and precise HPLC method for simultaneous determination of ALG and PGN in bulk powder and in their pharmaceutical dosage form (HPLC-2).

The new methods were developed without use of any buffer applying simple mobile phases with the advantage of easy preparation and enhanced resolution results and offering mobile phases that are suitable for both UV detection and mass spectrometric detection. The present work was proved to be more economic than most of the developed methods in the literature [4–11] as lower retention times ensures lower run time that decrease the required amount of mobile phase. Furthermore, the calculated resolution values from the present investigation showed satisfactory results that ensures good separation and accurate determination of the drugs.

## 2. Experimental

**2.1 Instrumentation.** The HPLC (10 AD, Shimadzu, Japan) consisted of a system controller (SCL-10A), degasser (DGU-12A), and UV–visible detector (SPD-10A) using XTERRA® C<sub>18</sub> column (250 mm × 4.6 mm, 5  $\mu\text{m}$ ), and software class VP and Elmasonic S 60 H degasser (Germany) were used.

**2.2 Reagents and Reference Samples.** EMG and LNG were found to be 99.71% and 100.21%, respectively, using reference method [4]; Glyxambi® tablets nominally containing 5 mg (LNG) and 10 mg (EMG) per tablet were supplied from Boehringer Ingelheim pharmaceutical company (Germany). ALG and PGN were found to be 99.87% and 100.03%, respectively, using reference method [10]; Oseni® tablets nominally containing 34 mg of alogliptin benzoate equivalent to 25 mg ALG and 33.06 mg of pioglitazone hydrochloride equivalent to 30 mg PGN per tablet were supplied from Takeda Pharmaceutical Company (Japan). HPLC-grade acetonitrile and methanol were purchased from Fisher Scientific (Loughborough, Leicestershire, UK). Orthophosphoric acid (85%) was purchased from VWR Chemicals (Pool, England). Formic acid was purchased from Sigma-Aldrich (Deisenhofen, Germany).

**2.3 Standard Stock Solutions.** Standard stock solutions of LNG, EMG, ALG, and PGN (1 mg mL<sup>-1</sup>) were prepared separately in methanol.

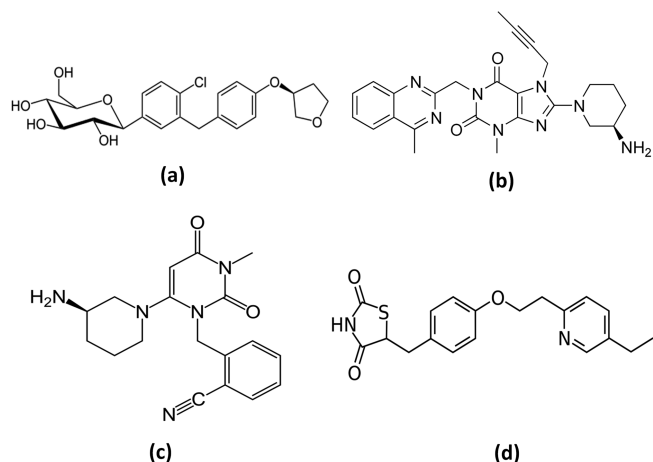
**2.4 Working Solutions.** All the working solutions were prepared separately using the mobile phase of each experiment. Solutions of LNG (100  $\mu\text{g mL}^{-1}$ ), EMG (200  $\mu\text{g mL}^{-1}$ ), ALG (50  $\mu\text{g mL}^{-1}$ ), and PGN (50  $\mu\text{g mL}^{-1}$ ) were prepared.

**2.5 Chromatographic Conditions.** All the mobile phases were filtered through 0.2- $\mu\text{m}$  membrane filter and degassed for 30 min in an ultrasonic bath prior to its use. The column temperature was kept at 25 °C, and the injection volume was 20  $\mu\text{L}$ . For HPLC-1, 0.1% aqueous formic acid–methanol–acetonitrile (40:20:40, v/v/v), pH 3.6, was used as a mobile phase at a flow rate of 2 mL min<sup>-1</sup> with UV detection at 226 nm. For HPLC-2, mobile phase consisting of 50% methanol at pH 2.7 (adjusted with orthophosphoric acid) was used at a flow rate of 0.8 mL min<sup>-1</sup> with UV detection at 270 nm.

**2.6 Sample Preparation.** Twenty tablets of Glyxambi® and Oseni® were separately weighed, powdered, and mixed in a mortar. Separately, accurately weighed amounts of the finely powdered Glyxambi® and Oseni® tablets equivalent to 20 mg

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**Figure 1.** Chemical structures of LNG (a), ALG (b), EMG (c), and PGN (d)

LNG and 40 mg EMG and 10 mg ALG and 9.7 mg PGN, respectively, were made up to 100 mL with methanol and sonicated to dissolve; then, the solutions were filtered.

## 2.7 Procedure and Method Validation

**2.7.1 Linearity.** Linearity was achieved over the concentration ranges of 2–50  $\mu\text{g mL}^{-1}$ , 4–100  $\mu\text{g mL}^{-1}$ , 0.5–25  $\mu\text{g mL}^{-1}$ , and 1–25  $\mu\text{g mL}^{-1}$  for LNG, EMG, ALG, and PGN, respectively, using LC–UV. Calibration curves were obtained by plotting peak area of six calibrators against concentration, and the regression equations were calculated.

**2.7.2 Assay of ALG, PGN, LNG, and EMG in bulk.** Concentrations equivalent to 5, 15, 25, 35, and 45  $\mu\text{g mL}^{-1}$  of LNG; 10, 30, 50, 70, and 90  $\mu\text{g mL}^{-1}$  of EMG; 2.5, 7.5, 12.5, 17.5, and 22.5  $\mu\text{g mL}^{-1}$  of ALG; and 2.5, 7.5, 12.5, 17.5, and 22.5  $\mu\text{g mL}^{-1}$  of PGN were prepared, and all the concentrations were calculated using regression equations.

**2.7.3 Precision.** Concentrations equivalent to 20, 25, and 30  $\mu\text{g mL}^{-1}$  LNG; 40, 50, and 60  $\mu\text{g mL}^{-1}$  EMG; and 8, 10, and 12  $\mu\text{g mL}^{-1}$  of both ALG and PGN were prepared and analyzed three times within the same day and on three successive days. The mean % recoveries and the relative standard deviations were calculated.

**2.7.4 Assay of LNG, EMG, ALG, and PGN in laboratory-prepared mixtures.** Different ratios (1:5, 2:5, 3:5,.....5:1) of LNG–EMG and ALG–PGN binary mixtures were prepared. Their concentrations were calculated using regression equations.

**2.7.5 Robustness.** Flow rate was changed by  $\pm 0.1$ , the organic strength was changed by  $\pm 2\%$ , and pH value of the mobile phase was varied by  $\pm 0.1$ .

**2.7.6 Assay of ALG, PGN, LNG, and EMG in Glyxambi® tablets and Oseni® tablets.** The sample solutions prepared under section Sample Preparation were diluted, and then, the procedures mentioned above were adopted. The concentrations of the mentioned drugs were calculated using their regression equations. Then, to check the validity of the proposed method, the standard addition technique was applied by adding different known concentrations of the pure drug to fixed concentrations of the drug product.

**2.7.7 Limit of detection and limit of quantitation.** Limit of detection ( $\text{LOD} = 3.3 \times \text{residual standard deviation of regression line / slope}$ ) and limit of quantitation ( $\text{LOQ} = 10 \times \text{residual standard deviation of regression line / slope}$ ) were determined for the proposed methods.

## 3. Results and Discussion

The proposed methods proved to be simple, accurate, and reproducible for the determination of LNG, EMG, ALG, and

PGN, and they were validated showing satisfactory data for all the parameters tested. The new methods were developed without buffer with the advantage of easy preparation and enhanced resolution results and offering mobile phases that are suitable for both UV detection and mass detection.

**3.1 Method Development.** According to the literature review regarding HPLC analysis of gliptins (LNG and ALG), Cyano column was used for chromatographic determination of single gliptins [12] while  $\text{C}_{18}$  column was used successfully for simultaneous determination of gliptins and other drugs in combination [13] with sharp peaks and good resolution, so  $\text{C}_{18}$  column was selected for the present work to ensure satisfactory separation of LNG–EMG peaks and ALG–PGN peaks.

For the first HPLC method, using both acetonitrile and methanol was crucial to enhance the resolution between LNG and EMG, and also, using 0.1% formic acid was enough to adjust the acidity of the mobile phase to pH 3.6. Flow rate was adjusted to be 2  $\text{mL min}^{-1}$  to decrease the retention time of the studied drugs without affecting the resolution between peaks. Finally, UV detection at 226 nm was the most sensitive wavelength for the determination of the two drugs simultaneously. Retention times for LNG and EMG were found to be 1.5 and 3 min, respectively, as shown in Figure 2.

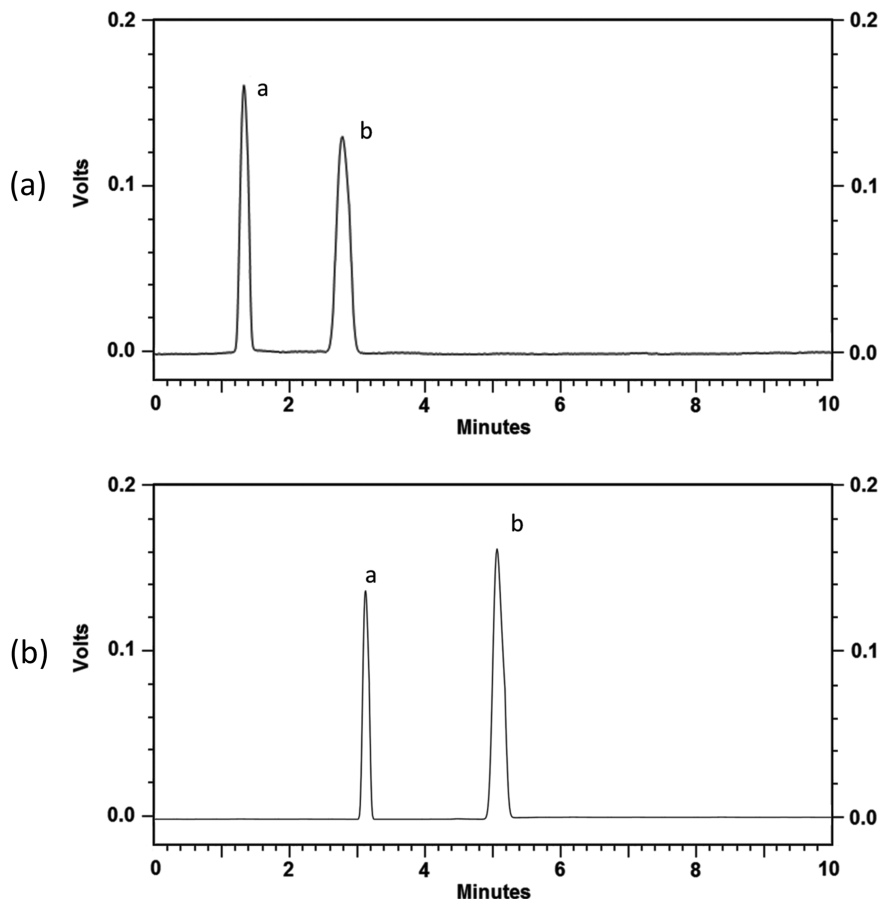
For the second HPLC method, the simplest mobile phase was developed for this method using 50% methanol and adjusting to the pH to 2.7 using orthophosphoric acid. The flow rate was set to be 0.8  $\text{mL min}^{-1}$  enhancing resolution between peaks by increasing the difference in their retention time. UV detection at the isosbestic point (270 nm) was the most sensitive wavelength for the determination of the two drugs simultaneously. Retention times for ALG and PGN were found to be 3.2 and 5.2 min, respectively, as shown in Figure 2.

**3.2 System Suitability Tests.** System suitability tests are an integral part of liquid chromatographic methods in the course of optimizing the conditions of the proposed method. System suitability tests are used to verify that the reproducibility was adequate for the analysis performance. The parameters of these tests are number of theoretical plates ( $N$ ), tailing of chromatographic peak ( $T$ ), capacity factor ( $K$ ), resolution between peaks ( $R_s$ ), and repeatability as percent relative standard deviation (% RSD) of peak area and retention time for six injections as shown in Table 1.

## 3.3 Methods' Validation According to ICH Guidelines [14]

**3.3.1 Linearity.** A linear relationship between peak area and component concentration was obtained for six chosen concentrations of each drug, and the regression equations were computed. The analytical data of the calibration curves are summarized in Table 2. The validity of the calibration curves was confirmed by the obtained low values of LOD–LOQ parameters and acceptable values of STEYX,  $S_b$ , and  $S_a$  as shown in Table 2, where LOD is the limit of detection which represents drug concentration at STEYX/ $S$  ratio of 3.3, LOQ is the limit of quantification at which STEYX/ $S$  is 10, STEYX is the residual standard deviation of the regression line,  $S_b$  is the standard deviation of the slope, and  $S_a$  is the standard deviation of the intercept.

**3.3.2 Accuracy.** Accuracy was calculated by % recovery of 5 different concentrations of each drug. The results including the mean of the recovery and standard deviation are shown in Table 3. In addition, accuracy was confirmed by % recovery of 9 different concentrations of each drug in the presence of the other drug in their laboratory-prepared mixtures. The results including the mean of the recovery and standard deviation are shown in Table 4. Furthermore, standard addition technique was applied to confirm and ensure the accuracy of



**Figure 2.** (a) LC chromatogram of (a) LNG (20 µg mL<sup>-1</sup>) and (b) EMG (40 µg mL<sup>-1</sup>) using UV detection at 226 nm; (b) LC chromatogram of (a) ALG (10 µg mL<sup>-1</sup>) and (b) PGN (9.7 µg mL<sup>-1</sup>), using UV detection at 270 nm

**Table 1.** System suitability tests for HPLC–UV methods for simultaneous determination of ALG and PGN and simultaneous determination of LNG and EMG

Item	ALG	PGN	LNG	EMG	Limits
<i>N</i>	1520	2809	3600	3580	>2000
<i>T</i>	1.02	1.04	1.05	1.04	≤2
<i>k</i>	3.00	4.80	1.20	2.60	≥1
<i>R<sub>s</sub></i>	5.26		5.13		>2
% RSD of 6 injections					
Peak area	0.16	0.29	0.30	0.15	≤2%
Retention time	0.23	0.14	0.26	0.18	≤2%

N indicates number of theoretical plates; T, tailing factor; K, capacity factor; *R<sub>s</sub>*, resolution; RSD, relative standard deviation.

the method and the concentrations were calculated using the corresponding regression equations as in Tables 5 and 6.

**3.3.3 Precision.** The % RSD values were calculated for the three concentrations of each drug mentioned under section Precision, within the same day and on three successive days, and found to be less than 1% as shown in Table 2.

**3.3.4 Specificity.** Specificity was confirmed by % recovery of 9 different concentrations of each drug in the presence of the other drug in their laboratory-prepared mixtures. The results including the mean of the recovery and standard deviation are shown in Table 4.

**3.3.5 Robustness.** There was no difference in the results obtained after small deliberate variations in some chromatographic

**Table 2.** Results of assay validation of HPLC–UV methods for simultaneous determination of ALG and PGN and simultaneous determination of LNG and EMG

Item	ALG	PGN	LNG	EMG
Range of linearity (µg mL <sup>-1</sup> )	0.5–25	1–25	2–50	4–100
Regression equation	Area × 10 <sup>-5</sup> = 0.8027C µg mL <sup>-1</sup> + 0.4424	Area × 10 <sup>-5</sup> = 0.7513C µg mL <sup>-1</sup> + 0.2048	Area × 10 <sup>-5</sup> = 1.4329C µg mL <sup>-1</sup> + 0.0213	Area × 10 <sup>-5</sup> = 0.5738C µg mL <sup>-1</sup> + 0.791
Regression coefficient ( <i>r</i> )	0.9999	0.9999	0.9998	0.9999
LOD (µg mL <sup>-1</sup> )	0.15	0.22	0.49	0.79
LOQ (µg mL <sup>-1</sup> )	0.47	0.68	1.50	2.40
<sup>a</sup> <i>S<sub>b</sub></i>	1.81 × 10 <sup>-3</sup>	2.50 × 10 <sup>-3</sup>	5.28 × 10 <sup>-3</sup>	1.69 × 10 <sup>-3</sup>
<sup>a</sup> <i>S<sub>a</sub></i>	0.034	0.046	0.20	0.13
Confidence limit of the slope	0.8027 ± 0.027	0.7513 ± 0.035	1.4329 ± 0.28	0.5738 ± 0.072
Confidence limit of the intercept	0.4424 ± 8.03 × 10 <sup>-4</sup>	0.2048 ± 5.11 × 10 <sup>-4</sup>	0.0213 ± 1.12 × 10 <sup>-4</sup>	0.791 ± 1.34 × 10 <sup>-4</sup>
Standard error of the estimation	0.037	0.051	0.21	0.14
Intraday precision % RSD	0.12 – 0.16 – 0.29	0.11 – 0.25 – 0.37	0.16 – 0.27 – 0.32	0.17 – 0.19 – 0.26
Interday precision % RSD	0.15 – 0.21 – 0.33	0.19 – 0.22 – 0.35	0.13 – 0.24 – 0.28	0.15 – 0.22 – 0.30
Robustness				
Flow rate (±0.1), % RSD	0.23	0.19	0.20	0.11
Organic strength (% ±2), % RSD	0.17	0.38	0.31	0.18
Mobile phase pH (±0.1), % RSD	0.28	0.14	NA	NA

<sup>a</sup>*S<sub>b</sub>* indicates standard error of intercept; *S<sub>a</sub>*, standard error of slope; NA: not applicable.

**Table 3.** Results for simultaneous determination of ALG and PGN and simultaneous determination of LNG and EMG in bulk powder by the two proposed HPLC–UV methods

ALG ( $\mu\text{g mL}^{-1}$ )		$^aR$ %	PGN ( $\mu\text{g mL}^{-1}$ )		$^aR$ %	LNG ( $\mu\text{g mL}^{-1}$ )		$^aR$ %	EMG ( $\mu\text{g mL}^{-1}$ )		$^aR$ %
Pure	Found		Pure	Found		Pure	Found		Pure	Found	
2.50	2.46	98.40	2.50	2.51	100.40	5.00	5.09	101.80	10.00	10.19	101.90
7.50	7.54	100.53	7.50	7.47	99.60	15.00	15.17	101.13	30.00	29.54	98.47
12.50	12.71	101.68	12.50	12.36	98.88	25.00	25.15	100.60	50.00	50.18	100.36
17.50	17.3	98.86	17.50	17.18	98.17	35.00	34.61	98.89	70.00	69.35	99.07
22.50	22.91	101.82	22.50	22.78	101.24	45.00	44.99	99.98	90.00	90.54	100.60
Mean $\pm$ SD		100.26 1.58	Mean $\pm$ SD		99.66 1.21	Mean $\pm$ SD		100.48 1.11	Mean $\pm$ SD		100.08 1.35

<sup>a</sup>Mean of three determinations.**Table 4.** Simultaneous determination of ALG and PGN and simultaneous determination of LNG and EMG by two HPLC–UV methods in laboratory-prepared mixtures

Ratio ALG–PGN	ALG ( $\mu\text{g mL}^{-1}$ )			PGN ( $\mu\text{g mL}^{-1}$ )			Ratio LNG–EMG	LNG ( $\mu\text{g mL}^{-1}$ )			EMG ( $\mu\text{g mL}^{-1}$ )			
	Pure	Found	$^aR$ %	Pure	Found	$^aR$ %		Pure	Found	$^aR$ %	Pure	Found	$^aR$ %	
5:1	25.00	25.50	102.00	5.00	4.91	98.20	5:1	50.00	50.09	100.18	10.00	10.09	100.90	
4:1	20.00	19.69	98.45	5.00	5.09	101.80	4:1	40.00	39.98	99.95	10.00	9.92	99.20	
3:1	15.00	14.80	98.67	5.00	4.97	99.40	3:1	30.00	29.87	99.57	10.00	10.17	101.70	
2:1	10.00	10.06	100.60	5.00	5.05	101.00	2:1	20.00	19.77	98.85	10.00	10.15	101.50	
1:1	5.00	5.05	101.00	5.00	5.07	101.40	1:1	10.00	10.20	102.00	10.00	9.97	99.70	
1:2	5.00	5.07	101.40	10.00	9.89	98.90	1:2	10.00	10.17	101.70	20.00	20.38	101.90	
1:3	5.00	5.08	101.60	15.00	15.07	100.47	1:3	10.00	10.06	100.60	30.00	29.54	98.47	
1:4	5.00	5.03	100.60	20.00	20.38	101.90	1:4	10.00	9.98	99.80	40.00	40.20	100.50	
1:5	5.00	4.94	98.80	25.00	25.06	100.24	1:5	10.00	10.12	101.20	50.00	49.92	99.84	
Mean $\pm$ SD		100.35 1.36			100.37 1.31			Mean $\pm$ SD		100.43 1.04			100.41 1.19	

<sup>a</sup>Mean of three determinations.**Table 5.** Simultaneous determination of LNG and EMG in pharmaceutical dosage form and standard addition technique by HPLC–UV method

Pharmaceutical dosage form	HPLC method											
	$^a\%$ Recovery $\pm$ SD		Standard addition technique									
	LNG	EMG	Claimed concentration ( $\mu\text{g mL}^{-1}$ )		Pure added ( $\mu\text{g mL}^{-1}$ )		Pure found		$^a\%$ R pure added			
		LNG	EMG	LNG	EMG	LNG	EMG	LNG	EMG			
Glyxambi® tablets (LNG and EMG) 5 mg/10 mg BN: 25061N					10.00	20.00	9.88	19.65	98.80	98.25		
					15.00	30.00	14.89	29.42	99.27	98.07		
			99.91 $\pm$ 0.83	100.25 $\pm$ 0.92	20.00	40.00	20.25	40.17	101.25	100.43		
					25.00	50.00	25.49	50.77	101.96	101.54		
					30.00	60.00	30.05	60.64	100.17	101.07		
				Mean $\pm$ SD				100.29 $\pm$ 1.32	99.87 $\pm$ 1.61			

<sup>a</sup>Mean of three determinations.**Table 6.** Simultaneous determination of ALG and PGN in pharmaceutical dosage form and standard addition technique by HPLC–UV method

Pharmaceutical dosage form	$^a\%$ Recovery $\pm$ SD		Standard addition technique									
	ALG	PGN	Claimed concentration ( $\mu\text{g mL}^{-1}$ )		Pure added ( $\mu\text{g mL}^{-1}$ )		Pure found		$^a\%$ R pure added			
			ALG	PGN	ALG	PGN	ALG	PGN	ALG	PGN		
Oseni® tablets (ALG and PGN) 34 mg/33.06 mg BN: 25059N					1.00	1.00	0.98	0.99	98.00	99.00		
					5.00	5.00	4.94	5.06	98.80	101.20		
			100.40 $\pm$ 1.51	100.31 $\pm$ 1.17	10.00	9.700	10.00	10.00	9.92	10.08	99.20	100.80
					12.5	12.50	12.69	12.71	101.52	101.68		
					15	15.00	15.30	14.81	102.00	98.73		
				Mean $\pm$ SD				99.90 $\pm$ 1.76	100.28 $\pm$ 1.33			

<sup>a</sup>Mean of three determinations.**Table 7.** Statistical comparison between the proposed methods and the reference methods

Statistical term	LNG		EMG		ALG		PGN	
	Reference method <sup>b</sup>	HPLC–UV method	Reference method <sup>b</sup>	HPLC–UV method	Reference method <sup>c</sup>	HPLC–UV method	Reference method <sup>c</sup>	HPLC–UV method
Mean	100.21	100.48	99.71	100.08	99.87	100.26	100.03	99.67
$\pm$ SD	1.29	1.11	0.97	1.35	1.30	1.58	1.54	1.21
% RSD	1.29	1.10	0.97	1.35	1.30	1.58	1.54	1.21
n	5	5	5	5	5	5	5	5
V	1.66	1.23	0.94	1.82	1.69	2.50	2.37	1.46
$t$ ( <sup>a</sup> 2.306)		0.36		0.50		0.43		0.41
$F$ ( <sup>a</sup> 6.39)		1.35		1.94		1.48		1.62

<sup>a</sup>Figures in parentheses are the theoretical  $t$  values at ( $p = 0.05$ ).<sup>b</sup>Reference method [6]: aliquots of standard solutions containing 10–50  $\mu\text{g mL}^{-1}$  LNG and 1–32  $\mu\text{g mL}^{-1}$  EMG were measured at 225 nm using LC–UV.<sup>c</sup>Reference method [12]: aliquots of standard solutions containing 5–25  $\mu\text{g mL}^{-1}$  ALG and 10–50  $\mu\text{g mL}^{-1}$  PGN were measured at 269 nm using LC–UV.

conditions as change in the flow rate, the organic strength, and the value of pH of the mobile phase as shown in Table 2.

**3.3.6 Pharmaceutical dosage forms and standard addition technique.** The proposed chromatographic methods were successfully applied to two pharmaceutical dosage forms. Standard addition technique was applied, and the concentrations were calculated using the corresponding regression equations as in Tables 5 and 6.

**3.3.7 Limit of detection and limit of quantification for the proposed method.** Limit of detection (LOD) and limit of quantitation (LOQ) were determined for the proposed methods, and results are given in (Table 2).

**3.4 Statistical Comparison.** Statistical comparison of the results obtained by the proposed methods and the reference methods [4 and 10] was carried out by “SPSS statistical package version 11” at  $P = 0.05$  as shown in Table 7.

#### 4. Conclusion

The proposed methods proved to be simple, accurate, and reproducible for the determination of LNG, EMG, ALG, and PGN, and they were validated showing satisfactory data for all the parameters tested. The developed methods can be conveniently used by quality control laboratories. The new

methods were developed without use of any buffer applying simple mobile phases with the advantage of easy preparation and more sensitive results and offering mobile phases that are suitable for both UV detection and mass detection.

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