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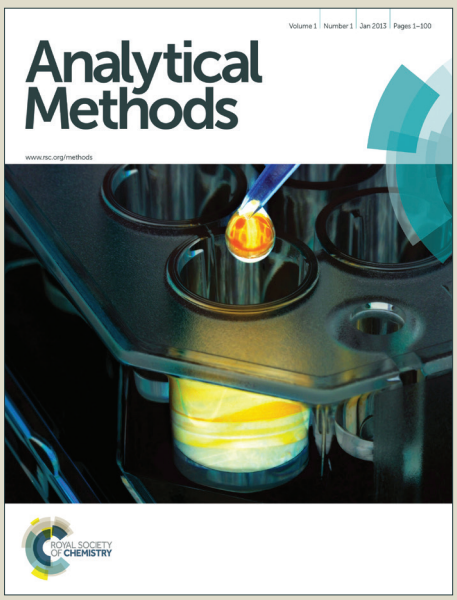

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Simultaneous Determination of Amlodipine besylate and Valsartan using Micelle-enhanced First Derivative Synchronous Spectrofluorimetric Method and Application in Their Co-formulated Tablets

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Abstract

A novel, highly sensitive and simple first derivative synchronous spectrofluorimetric method was developed for simultaneous determination of the binary mixture of amlodipine besylate (AML) and valsartan (VAL) in their co-formulated tablets. The proposed method is based on measurement of the synchronous fluorescence intensity of these drugs at $\Delta\lambda$= 80 in aqueous sodium dodecyl sulphate (SDS) system. The fluorescence intensities of both AML and VAL were greatly enhanced (200% and 220 % for AML amd VAL respectively) in the presence of SDS. The different experimental parameters affecting the fluorescence of the two drugs were carefully studied and optimized. The fluorescence-concentration plots were rectilinear over the range of 0.5–4 μg/mL and 0.05–3.0 μg/mL for VAL and AML, respectively with lower detection limits (LOD) of 0.027 and 0.022 μg/mL and quantification limits (LOQ) of 0.083 and 0.007 μg/mL for VAL and AML, respectively. The proposed method was successfully applied for the determination of the two compounds in laboratory prepared mixtures and in commercial tablets. The proposed method was successfully applied to the content uniformity testing of tablets.

Key words: Micelle, spectrofluorimetry, synchronous, valsartan, amlodipine tablet dosage forms and content uniformity testing.

Inroduction

Valsartan, (VAL) (S)-N-(1-Oxopentyl)-N-[[2'-(1H-tetrazol-5- yl) [1,1'-biphenyl]-4-yl]methyl]-L-valine (Fig. 1), is an orally active specific angiotensin II receptor blocker effective in lowering blood pressure in hypertensive patients. It is a selective type-1 angiotensin II receptor antagonist which blocks the blood pressure increasing effects of angiotensin II via rennin-angiotensin-aldosterone system. It is used as a first line agent to treat uncomplicated hypertension, isolated systolic hypertension and left ventricular hypertrophy[1].

Amlodipine besylate (AML) chemically known as 3-ethyl-5-methyl (4R,S)-2-[(2-aminoethoxy) methyl]-4-(2-chlorophenyl)-6-methyl-1,4-dihydropyridine 3,5-dicarboxylate benzenesulphonate (Fig. 1) is a calcium channel blocker that inhibits the transmembrane influx of calcium ions into vascular smooth muscle and cardiac muscle and is used in the treatment of hypertension and angina. [2]

Fast and reliable simultaneous determination of AML or VAL in pharmaceutical dosage forms is required due to the therapeutic importance. There are a few methods including spectrophotometry [3,4], potentiometry [5], thin-layer chromatography [6], HPLC [6-12], and capillary electrophoresis [13] reported for the determination of the assay of this combination. However, these methods are usually laborious, expensive, time-consuming and complex to be operated.

Spectrophotometrially, Shaalan and Belal [14] described a method for the determination of the two drugs in their combined tablets. The method involved the measurement of the native fluorescence of the two drugs at 360/455 nm in distilled water and at 245/ 378 nm in 0.1M acetic acid for AML and VAL repectively, The method does not allow the simultaneous determination of the two compounds stated

by the authors. Therefore, a fast, simple, low cost, accurate, precise and sensitive method is very important especially for routine simultaneous determination of pharmaceuticals containing both AML and VAL.

The aim of the present study is to establish and develop a novel, sensitive and selective derivative synchronous fluorescence spectroscopic (DSFS) method for simultaneous determination of AML and VAL either per se or in pharmaceutical preparations. The normal synchronous fluorescence spectra of AML and VAL are greatly overlapped. Such problem encouraged us to utilize a simple first derivative synchronous fluorescence spectroscopy (FDSFS) to solve such problem through measuring peak intensities at 362 nm and 300 nm for AML and VAL respectively. The developed method was applied for the simultaneous determination of AML and VAL in their co-formulated pharmaceutical preparation.

Synchronous fluorescence spectroscopy (SFS) has several advantages over conventional fluorescence spectroscopy, including simple spectra, high selectivity and low interference [15]. Because of its sharp, narrow spectrum, SFS serves as a very simple, effective method of obtaining data for quantitative determination in a single measurement [16]. The combination of SFS and derivatives is more advantageous than the conventional emission spectrum in terms of sensitivity, because the amplitude of the derivative signal is inversely proportional to the band width of the original spectrum [17-18].

Recently, derivative synchronous fluorometry (DSF) has been utilized for determination of several mixtures in their co-formulated dosage forms and biological fluids. Mixtures of sulpiride and mebeverine[19], cinnarizine and domperidone [20], metoclopramide and pyridoxine [21], aspirin with salicylic acid [22], diflunisal and salicylic acid [23], carvedilol and ampicillin [24], sulpirid and its alkaline degradation [25] and ethamsylate [26] have been determined through this approach.

EXPERIMENTAL

Material

AML and VAL pure samples were purchased from Sigma (St. Louis, Mo, USA). Exforge® tablets (Batch no. Y0001/50002) (product of Novartis, from local pharmacy in Egypt) were used as pharmaceutical dosage form which contains 160 mg of VAL and 10 mg AML per tablet.

Reagents

All reagents and solvents were of Analytical Reagent Grade, Methanol (Merck, Darmstadt, Germany). Acetate buffer 0.2 M (pH3.6-5.6) was prepared by mixing appropriate volume of 0.2 M acetic acid with 0.2 M sodium acetate. Borate buffers (pH 5.5-13) were prepared by mixing appropriate volumes of 0.02 M boric acid with 0.2 M sodium hydroxide, the pH was adjusted to the required pH using pH meter, 1% aqueous SDS solution, β-cyclodextrin (β-CD) and hydroxy propyl-β-cyclodextrin (HP-β-CD) were obtained from Merck (Germany), cetyl trimethyl ammonium bromide (CTAB) was purchased from Winlab (UK) , Tween-80 and methyl cellulose were obtained from El-Nasr Pharmaceutical Chemical Co. (ADWIC; Egypt).

Apparatus

- Fluorescence spectra and measurements were recorded using a Perkin -Elmer UK model LS 45 luminescence spectrometer, equipped with a 150 Watt Xenon arc lamp, grating excitation and emission monochromators for all measurements and a Perkin-Elmer recorder. Slit widths for both monochromators were set at 10 nm. A 1 cm quartz cell was used. Derivative spectra can be evaluated using Fluorescence Data Manger (FLDM) software.

For best resolution and smoothing, number of points of 99 was used for deriving the first derivative spectra. The fluorescence intensities of the first derivative spectra were estimated at 362 and 300 nm for AML and VAL, respectively.

- A pH Meter (Model pHS-3C, Shanghai Leici instruments Factory, China) was used for pH adjustment.
- Sonicator BHA-180T (Abbotta Corporation, USA) was used.

Standard solutions

Stock solutions of 100 µg /mLof AML or VAL were prepared in methanol. Working solutions were prepared by appropriate dilution of the stock solutions with dist. water. The stock solution was found to be stable for 1 week if kept in the refrigerator.

General procedures of the calibration curves:

Aliquot volumes of the working solutions over concentration ranges 0.05–3.0 µg/mL of AML or 0.5– 4.0µg/mL of VAL were transferred into a series of 10 mL volumetric flasks. 0.5 mL of 1.0 % SDS was added and the solutions were diluted to the volume with dist. water and mixed well. Synchronous fluorescence spectra of the solutions were recorded by scanning both monochromators at a constant wavelength difference $\Delta\lambda$ =80 nm and scan rate of 600 nm min^{-1} using 10 nm excitation and emission windows. The first derivative fluorescence spectra of VAL and AML were derived from the normal synchronous spectra using FLDM software. The peak amplitude of the first derivative spectra was estimated at 362 nm and 300 nm for AML and VAL, respectively. A blank experiment was performed simultaneously. The peak amplitude of the first derivative technique was plotted versus the final concentration of the drug (µg/mL) to get the calibration graph. Alternatively, the corresponding regression equations were derived.

Procedure for the laboratory prepared mixture:

Aliquot volumes of AML and VAL standard solutions in the pharmaceutical ratio of 1:16 were transferred into a series of 10 mL volumetric flasks. 0.5 mL of 1% SDS solution was added and diluted to the volume with distilled water, and mixed well. The recommended procedure described under calibration curve was then performed. The peak amplitude of the first derivative technique was plotted vs the final concentration of the drug (µg/mL) to generate the calibration graph. Alternatively, the corresponding regression equations were derived.

Procedure for commercial tablets:

Ten tablets (Exforge® tablets) were weighed and pulverized well. A weighed quantity of the powdered tablet equivalent to 160 mg VAL and 10 mg of AML (in their pharmaceutical ratio of 16:1) was transferred into a small conical flask and extracted with 3 x 30 mL of methanol. The extract was filtered into a 100 mL volumetric flask. The conical flask was washed with few milliliters of methanol. The washings were passed into the same volumetric flask and completed to the volume with the same solvent. Aliquots covering the working concentration range were transferred into 10 mL volumetric flasks. The recommended procedure under "Calibration Curve" was performed. The nominal content of the tablets were determined either from a previously plotted calibration graph or using the corresponding regression equation.

Content uniformity testing:

The same procedure applied for the analysis of AML and VAL in their tablets was followed using one tablet as a sample. Ten tablets were analyzed and the uniformity of their contents was tested by applying the official USP [27] guidelines.

RESULTS AND DISCUSSION:

Both of AML and VAL exhibit enhanced fluorescence at 438 nm and 374 nm, after excitation at 237 nm for AML and VAL respectively (Fig. 2). Both the excitation and emission spectra of AML and VAL overlapped (Fig. 2). This fact hindered the use of this method for the simultaneous determination of AML and VAL. This problem is aggravated if it is desired to determine these compounds in their co-formulated preparations. It was necessary to record first, the normal synchronous spectra for AML and VAL in order to derive the first derivative synchronous spectra. Fig. 3a, shows the SF spectra of different concentrations of VAL at 284 nm in presence of constant concentration AML (2.0 μg/mL), whereas Fig. 3b, illustrates the SF spectra of different concentrations of AML at 359 nm in presence of constant concentration of VAL (2.0 μg/mL).

Therefore the first derivative synchronous fluorescence spectroscopy (FDSFS) technique was chosen for simultaneous determination of both of AML and VAL in their tablets. Spectra of AML and VAL were well separated using FDSFS with a zero-crossing technique of measurement (Figs. 4a and b). Under the experimental conditions the two peaks appeared at 363 and 300 nm for AML and VAL respectively.

Optimization of Reaction condition

Different experimental parameters affecting the performance of the proposed method were carefully studied and optimized. Such factors were changed individually while others were kept constant. We aimed to enhance the emission spectra of AML and VAL in order to explore a new methodology for the analysis of these drugs in different pharmaceutical preparation. It is well known that the addition of a surfactant at a concentration above its critical micellar concentration to a given fluorophore solution increases the molar absorbtivity and\or the fluorescence quantum yield of the fluorophore in many cases [28, 29]. This fact has been used to improve the performance of the proposed method. The fluorescence properties of AML and VAL

in various micellar media were studied; there was an enhancement (about 300%) of the fluorescence intensity in the presence of SDS compared with aqueous solution.

Selection of optimum Δλ:

The optimum Δλ value is important for performing the synchronous fluorescence scanning technique with regards to its resolution, sensitivity and features. It can directly influence synchronous spectral shape, band width and signal value. For this reason a wide range of Δλ (20, 40, 60, 80, 100 and 120 nm) was examined. When Δλ was less than 80 nm, the spectra shapes were irregular and noisy with weak fluorescence intensities. On the other hand, when Δλ was more than 80 nm, overlapping of the two peaks with poor separation was achieved. Therefore, Δλ of 80 was chosen as optimal for separation of AML and VAL mixtures, since it resulted in two distinct peaks with good regular shapes and reduced the spectral interference caused by each compound in the mixture.

Selection of optimum pH:

The influence of pH on the synchronous fluorescence intensities of the two drugs was studied using different buffers covering the whole pH range, e.g. acetate buffer over the pH range of 3.6-5.6 and borate buffer over the pH range 5.5-13. The synchronous fluorescence intensity of AML and VAL is not affected upon increasing the pH values up to 8 and further increase in pH resulted in a gradual decrease in the synchronous fluorescence intensities, after which it extremely decreased at pH 13 (Fig. 5). Therefore, no buffer was used throughout the study.

Selection of organized media:

The fluorescence intensities of AML and VAL in various organized media were studied using anionic surfactant (SDS), cationic surfactant surfactant (CTAB) non-ionic surfactant (Tween-80) and different macro molecules such as, methyl cellulose and ß- CD. For AML and VAL, CTAB, methyl cellulose and ß- CD has no effect on relative fluorescence intensities (RFI) of the studied drugs, while Tween-80 caused a slight decrease in its RFI. Only SDS gave a considerable increase in the RFI so, SDS was selected as the fluorescence enhancer for both drugs (Fig. 6).

Effect of the volume of SDS:

The influence of volume of SDS on the fluorescence intenisty was studied using increasing volumes of 1% SDS. It was found that increasing volumes of SDS solution resulted in a gradual increase in the fluorescence intensities up to 0.5 mL after which further increase in volume produced no further increase in RFI. So, 0.5 mL 1% w\v SDS solution was chosen as the optimum volume for both AML and VAL.

Effect of diluting solvent

The effect of different diluting solvents on the RFI of AML and VAL in the presence of SDS was investigated using water, methanol, acetonitrile, n-propanol, dimethyl sulphoxide and dimethyl formamide. It was found that water was the optimum solvent for dilution, as it gave the highest RFI and the lowest blank reading. Distinct and sharp decrease in the relative fluorescence intensities was achieved in the SDS system using methanol, acetonitrile or n-propanol. This effect is attributed to their denaturating effect on the micelles, where short-chain alcohols (methanol and propanol) are solubilized mainly in the aqueous phase and affect the micellization process by modifying the solvent properties. Addition of these organic solvents also results in a reduction of the size of the micelles, but with a progressive breakdown of the surfactant aggregate at very high concentration [30]. Both dimethyl formamide and dimethyl sulphoxide quenched the fluorescence intensities of AML and VAL, since they initiated an intersystem crossing process (similar to the heavy atom effect) [31].

Analytical Performance

The first derivative synchronous fluorescence spectroscopy (FDSFS) - concentration plots for the two drugs were linear over the concentration range showed in Table 1. Linear regression analysis of the data gave the following equations:

$D^1 = 0.88 + 58.46C$ (r=0.9999), for AML at 362 nm.

$D^1 = -9.95 + 42.8C$ (r=0.9999), for VAL at 300 nm.

Where D^1 is the first derivative synchronous fluorescence spectroscopy, C is the concentration of the drug (μg/mL) and r is correlation coefficient. The limits of quantification (LOQ) was calculated according to ICH Q2B recommendations [32],.

The limits of detection (LOD) was also calculated according to ICH Q2B recommendations [32]. The results of LOD and LOQ of AML and VAL respectively are abridged in Table 1.

LOQ and LOD were calculated according to the following equations [32]: LOQ=10 σ/S

LOD=3.3 σ/S

Where, σ is the standard deviation of the intercept of regression line and S is the slope of regression line of the calibration curve. The proposed method was evaluated by studying the accuracy as percent relative error and precision as percent relative standard deviation. The results are abridged in Table 1. Statistical analysis [33] of the results, obtained by the proposed and the reference method [14] using Student's t-test and variance ratio F-test, shows no significant difference between the performance of the two methods regarding the accuracy and precision, respectively (Table 2).

Analysis of laboratory prepared mixture of AML and VAL:

The proposed method was applied for the simultaneous determination of AML and VAL in their laboratory prepared mixtures of both drugs in their pharmaceutical ratio of 1 :16. The relative fluorescence intensities of first derivative technique were measured for both drugs. The first derivative synchoronous spectrum of AML was measured at 362 nm which is considered as zero crossing point for VAL and the first derivative signal for VAL was measured at 300 nm which is the zero crossing point for AML. The concentrations of both drugs in the laboratory prepared mixtures were calculated according to their linear regression equation of the calibration graphs. The results indicate high accuracy of the proposed method as shown in Table 3.

Accuracy and Precision

Repeatability

The repeatability was evaluated by applying the proposed method for the determination of three concentrations of AML and VAL in pure forms on three successive times, and the results are illustrated in Table 4. The low %Error and low % RSD indicates high accuracy and high precision of the proposed method respectively.

Intermediate precision

Intermediate precision was performed through replicate analysis of AML and VAL in pure form. The results are shown in Table 4, for a period of three successive days.

Robustness of the method

The robustness of the proposed method is demonstrated by the constancy of the synchronous fluorescence intensities with the deliberated changes in the experimental parameters such as change volume of SDS 0.5 ml ± 0.1. This minor change that may take place during the experimental operation didn't greatly affect the fluorescence intensity of the AML and VAL.

Pharmaceutical Applications

Selectivity

The proposed method was applied for the determination of AML and VAL in their co-formulated tablets. The specificity of the method was investigated by observing any interference encountered from the common tablet excepients, such as lactose, gelatin, magnesium stearate and starch. These excepients did not interfere with the proposed method (Table 5).

Content uniformity testing

Due to high precision of the proposed method and its suitability for analysis of AML and VAL in their dosage forms with sufficient accuracy, the method is ideally suited for content uniformity testing which is time consuming process when using

conventional assay techniques. The steps of the test were adopted according to the commercially available tablets and it was found to be smaller than the maximum allowed acceptance value (L1). The results demonstrated good drug uniformity as shown in (table 6).

CONCLUSION:

A new simple and sensitive method was explored for the simultaneous determination of VAL and AML in their co-formulated tablets. The first derivative synchronous spectrofluorimetric method, by virtue of its high sensitivity, could be applied to the analysis of both drugs in their co-formulated dosage forms with low detection limit (LOD) =0.027 and 0.007 μg/mL for VAL and AML respectively. In addition, a simple sample preparation enables the use of this method for routine quality control of VAL and AML in commercial tablets with good accuracy and excellent application of content uniformity test.

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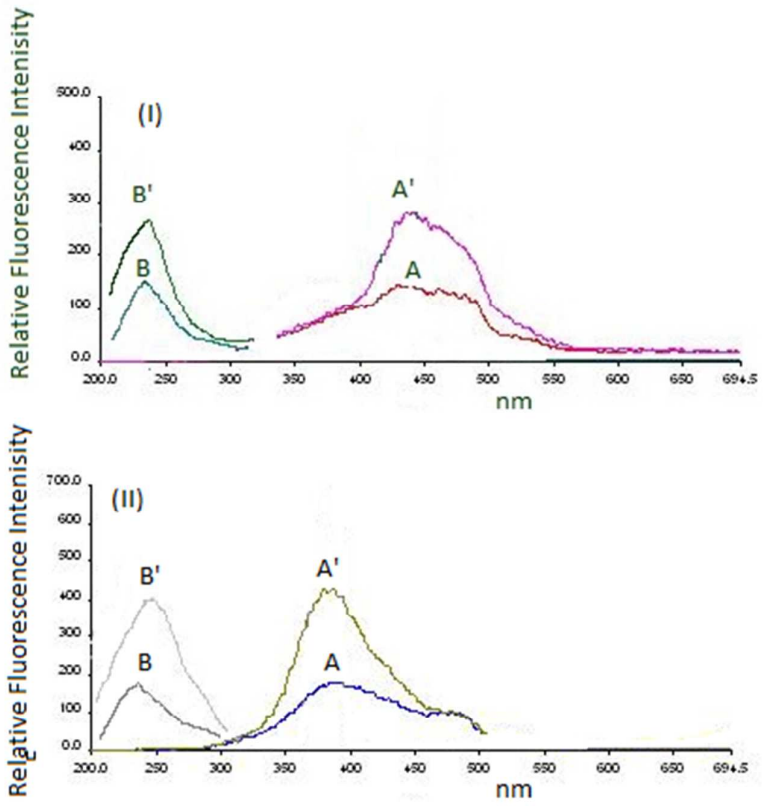


Fluorescence spectra: (I), A and B are emission and excitation spectra of AML (2 μg/ml) in methanol while A' and B' are emission and excitation spectra of AML (2 μg/ml) in SDS system.

(II), A and B are emission and excitation spectra of VAL (2 μg/ml) in methanol while A' and B' are emission and excitation spectra of VAL (2 μg/ml) in SDS system.

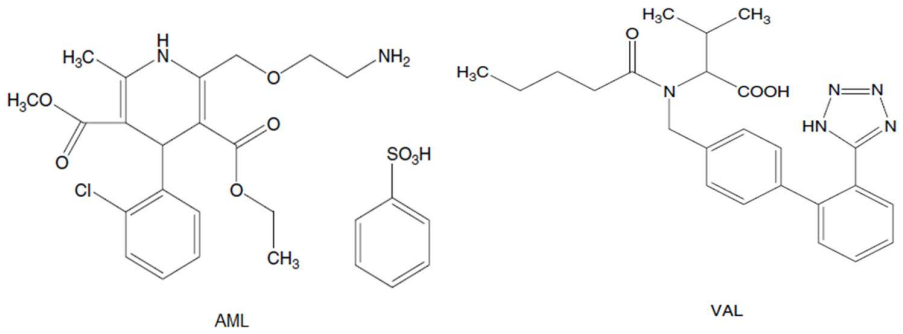


Figure 1 : Structural formula of amlodipine besylate and valsartan

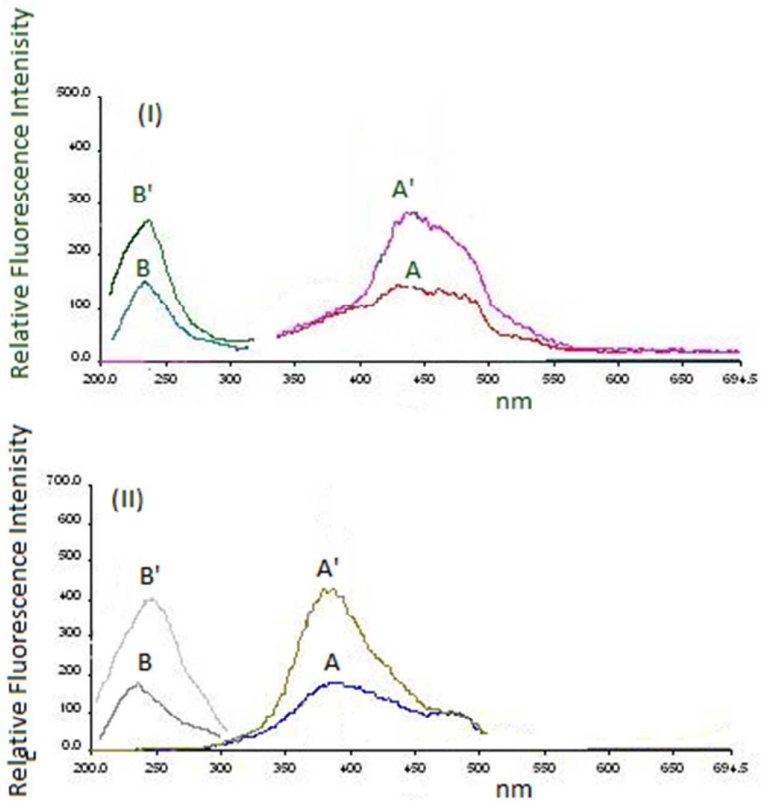
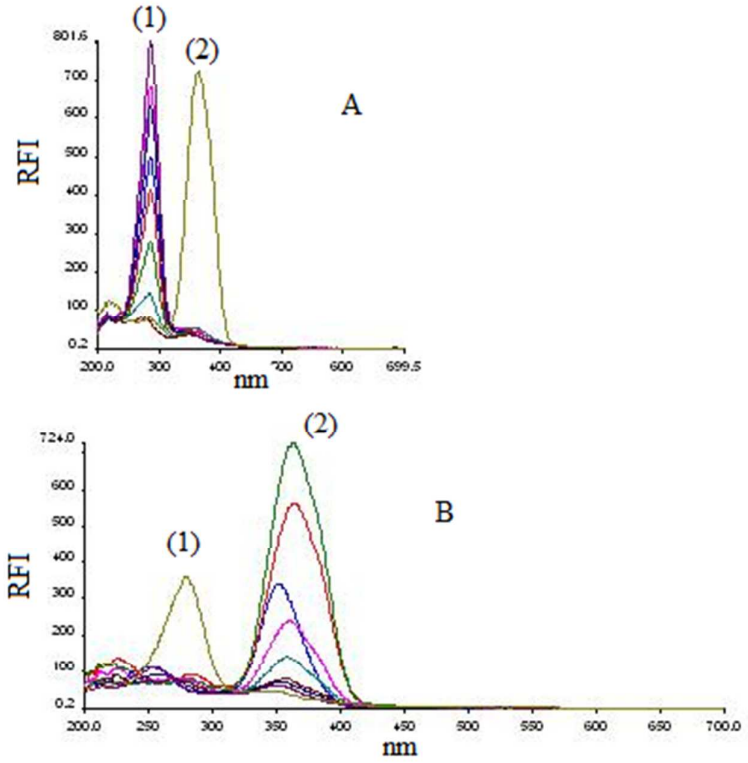


Figure 2: Fluorescence spectra: (I), A and B are emission and excitation spectra of AML (2 µg/ml) in methanol while A' and B' are emission and excitation spectra of AML (2 µg/ml) in aqueous SDS system.

(II), A and B are emission and excitation spectra of VAL (2 µg/ml) in methanol while A' and B' are emission and excitation spectra of VAL (2 µg/ml) in aqueous SDS system.



Figuer 3: (A) Synchronous fluorescence spectra of VAL at 284 nm and AML. (1) a-f spectra of VAL(0.5–4.0 μg/ml); (2), spectrum of AML. (B) Synchronous fluorescence spectra of AML at 359 nm and VAL.(2) a-f spectra of AML (0.05-3.0 μg/ml); (1) spectrum of VAL

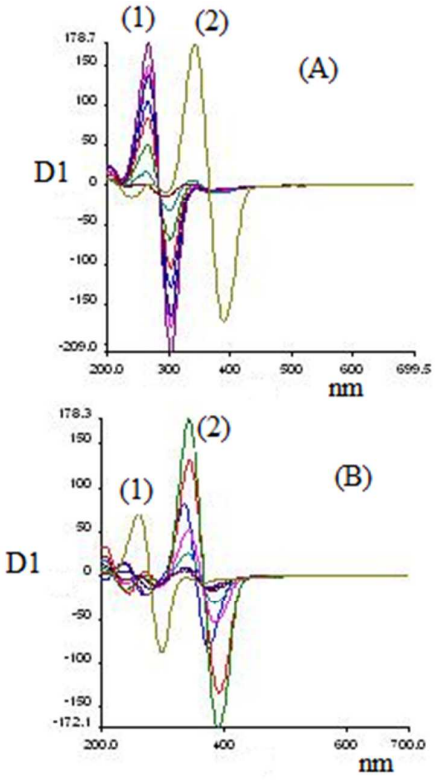
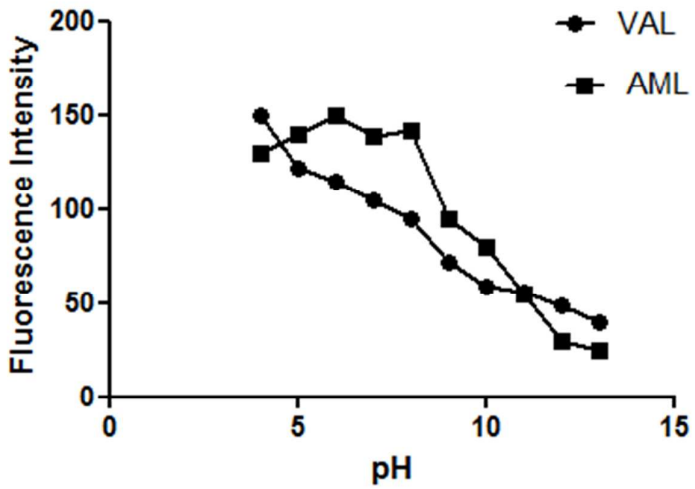


Figure 4: (A) First derivative synchronous fluorescence spectra of VAL at 300 nm and AML, (1), a–f Spectra of VAL (0.5–4.0 µg/ml) ; (2), Spectrum of AML (2 µg/ml) respectively.

(B) First derivative synchronous fluorescence spectra of AML at 362 nm and VAL, (2), a–f Spectra of AML (0.05–3.0 µg/ml) ; (1), Spectrum of VAL (2 µg/ml) respectively.



Figuer 5: Effect of pH on fluorescence intensity of VAL (1 µg/mL) and AML (1 µg/mL).

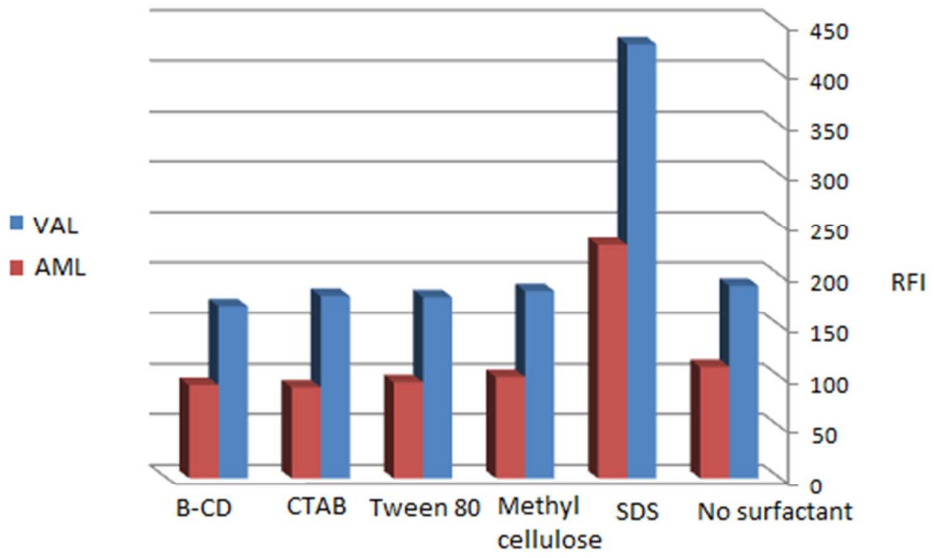


Figure 6: Effect of the organized media (0.5 mL 1% solution of each) on RFI of VAL (2 µg/mL) and AML (2 µg/mL).

Table 1: Analytical performance data for the proposed method

Parameter	AML	VAL
Concentration range (µg/mL)	0.05-3.0	0.5- 4.0
Limit of detection (LOD) (µg/mL)	0.007	0.027
Limit of quantification (LOQ) (µg/mL)	0.022	0.083
Correlation coefficient (r)	0.9999	0.9999
Slope	58.46	42.8
Intercept	0.88	-9.95
Standard deviation of the residuals ($S_{y/x}$)	0.260	0.423
Standard deviation of the intercept (S_a)	0.134	0.359
Standard deviation of the slope (S_b)	0.0933	0.134
RSD (%)	0.755	0.328
Error (%)	0.266	0.134

Table 2: Application of the proposed method and reference method for the determination of AML and VAL in pure form:

Parameter	Proposed method			Reference method [14]	
	Conc. taken (µg/mL)	Conc. found (µg/mL)	Recovery %	Conc. taken (µg/mL)	Recovery %
AML	0.05	0.05	99.69	1.00	99.17
	0.10	0.10	98.74	2.00	101.19
	0.25	0.25	99.00	3.00	99.54
	0.50	0.50	99.27		
	1.00	1.01	100.76		
	1.50	1.50	100.13		
	2.00	2.00	100.16		
	3.00	3.00	99.84		
$\bar{X}$ ±SD			99.59 ± 0.752		99.96 ± 1.07
t			0.724 (1.833)*		
F			2.01 (4.74)*		
VAL	0.50	0.50	100.15	0.01	99.55
	1.00	1.00	100.24	0.02	100.40
	2.00	2.01	100.27	0.03	99.87
	3.00	2.98	99.44		
	3.50	3.50	99.93		
	4.00	4.01	100.29		
$\bar{X}$ ±SD			100.05 ± 0.328		99.94 ± 0.427
t			0.462 (1.833)*		
F			1.703 (4.74)*		

- * The values of tabulated t and F at p = 0.05
- Each result is the average of three separate determinations.

Table3. Application of the proposed method for determination of AML and VAL in laboratory prepared mixture

Sample	Concentration taken (µg/ml)		Concentration found (µg/ml)		Recovery %	
	AML	VAL	AML	VAL	AML	VAL
AML and VAL mixture	0.05	0.80	0.05	0.81	100.72	100.92
	0.10	1.60	0.10	1.59	99.46	99.32
	0.20	3.20	0.20	3.20	100.09	100.11
X`					100.09	100.11
± S.D.					±0.629	±0.795
%RSD					0.628	0.794
%Error					0.363	0.0.459

-Each result is the average of three separate determinations

Table 4: Accuracy and precision for the determination of AML and VAL using the proposed method.

Drug	Conc. taken (µg/mL)	Intraday percision			Interday precision		
		Found (%)*	RSD %	Error %	Found (%)*	RSD %	Error %
AML	0.05	99.72± 0.339	0.340	0.196	100.28 ± 0.205	0.205	0.118
	0.10	99.73 ± 0.643	0.642	0.370	98.69 ± 0.391	0.390	0.225
	0.20	99.31 ± 0.633	0.634	0.366	99.58 ± 0.578	0.578	0.333
VAL	0.80	100.25 ±0.274	0.275	0.158	99.39 ± 0.205	0.205	0.118
	1.60	99.35 ± 0.506	0.506	0.292	98.69 ± 0.391	0.390	0.225
	3.20	100.20 ± 0.591	0.591	0.341	99.58 ± 0.574	0.575	0.331

-*Each result is the average of three separate determinations.

Table 5. Application of the proposed method for determination of AML and VAL in their co-formulated preparations

Preparation	Concentration taken (µg/ml)		Concentration found (µg/ml)		Recovery %	
	AML	VAL	AML	VAL	AML	VAL
Exforge® tabletsa	0.05	0.80	0.05	0.81	100.72	101.58
(VAL 160mg +AML 10mg/ tab	0.10	1.60	0.10	1.58	99.32	98.84
(Batch no. Y0001/50002)	0.20	3.20	0.20	3.21	100.13	100.19
X`					100.05	100.20
± S.D.					±0.702	±1.366
%RSD					0.702	1.364
%Error					0.405	0.0.789

-Each result is the average of three separate determinations.

-[a] product of Novartis, Pharma (Basel, Switzerland) from local pharmacy in Egypt.

Table 6. Content uniformity testing of AML and VAL in co-formulated tablets using the proposed method

Parameter	Percentage of the label claim in Exforge® tablets	
	AML	VAL
Data	100.00	106.30
	108.48	98.08
	102.42	99.78
	96.36	100.63
	98.18	101.62
	101.21	98.65
	92.12	103.18
	95.15	100.84
	95.75	101.20
	97.57	99.78
$\bar{X}$	98.72	101.00
%RSD	4.60	2.30
%Error	1.46	0.73
Acceptance value (AV) [27]	10.99	5.66
Max. allowed AV (L1) [27]	15.00	