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Spectrophotometric and High Performance Liquid Chromatographic Methods for the Determination of Dapsone in a Pharmaceutical Preparation

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ABSTRACT

The detailed investigation of a sensitive and selective spectrophotometric method for the determination of dapsone; based on the oxidative coupling reaction of dapsone drug with pyrocatechol in the presence of potassium periodate in acidic medium to form an intense, stable, purple-red coloured water-soluble dye, which shows a maximum absorption at 509 nm. Beer's law obeyed over the range of 10-500 µg /25 ml, i.e,0.4-20 ppm with a molar absorptivity of 1.05×10^{4}l.mol^{-1}.cm^{-1}and Sandell's sensitivity index of 0.0235 µg.cm^{-2},a relative standard deviation of ±2.60 to ±1.05%; and limit of detection, (LOD) of 0.1735 µg.ml^{-1}.; and (LOQ) of 0.5785µg.ml^{-1}. And the method does not require temperature control or solvent extraction. HPLC technique has been also developed for the measurement of dapsone. The analysis was achieved on a C_{18} column using water and acetonitrile in the ratio of 50:50 (V:V) as a mobile phase at a flow rate equals to 0.5 ml / min. and the detection was done spectrophotometrically at 297 nm. A linear relationship is obeyed over the range 0.5-20 ppm with a relative standard deviation (RSD) ±1.53 to ±0.83% and relative error -0.3 to +1.1 %. Both methods were applied successfully to the assay of dapsone in pharmaceutical preparation (Tablet).

Keywords: Spectrophotometricy, high performance liquid chromatography, dapsone, pyrocatechol, reagent.

التقدير الطيفي والكروماتوغرافي السائل عالي الاداء لتقدير الدابسون في مستحضر دوائي

الملخص

يتضمن البحث طريقة طيفية حساسة وانتقائية لتقدير الدابسون والتي تعتمد على مفاعلة الدابسون مع الكاشف (البايروكاتيكول) بوجود عامل مؤكسد (بيرايوديد البوتاسيوم) في وسط حامضي لتكوين صبغة ثابتة وذائبة في الماء ذات لون وردي – محمر وتعطي اعلى امتصاص عند الطول الموجي 509 نانوميتر، وكانت حدود قانون بير بين 10-500 مايكروغرام / 25 مللتر (0.4-20 جزء بالمليون).

وكانت الامتصاصية المولاري 1.05×10^4 لتر. مول$^{-1}$.سم$^{-1}$. ودلالة ساندل 0.0235 مايكروغرام. سم$^{-2}$. والخطأ النسبي بين ±2.60 و 1.05± % اعتمادا على مستوى التركيز للادواء مما يدل على ان الطريقة تمتاز بدقة وتوافق عاليين. وبحد كشف 0.1735 مايكروغرام / مللتر وحد تقدير كمي 0.5785 مايكروغرام / مللتر. ومن محاسن الطريقة انها لا تحتاج الى السيطرة على درجة الحرارة ولا الى عملية استخلاص بالمذيب. اما الطريقة الثانية فتتضمن حقن الدابسون الى عمود HPLC نوع C_{18} باستخدام مزيج من الماء : الاسيتونتريل بنسب 50:50 (V:V) بوصفه طوراً متحركاً وبمعدل سرعة جريان 0.5 مللتر للدقيقة الواحدة. وأستخدم الطول الموجي 297 نانوميتر في الكشف والتقدير الكمي. تتبع الطريقة حدود تقديرية من 0.5 الى 20 جزء لكل مليون. إذ تراوح الانحراف القياسي النسبي بين ±1.53 و±0.83 % والخطأ النسبي بين 0.3 – و1.1+%. وقد تم تطبيق كلا الطريقتين وبنجاح لتقدير الدابسون في الاقراص الدوائية.

الكلمات الدالة: طيف الامتصاص، كروماتوغرافي السائل عالي الأداء، الدابسون، البايروكاتيكول.

INTRODUCTION

Dapsone (diamino-diphenyl sulfone, DDS), is a white or slightly yellowish-white crystalline powder. It discolours on exposure to light but this is not accompanied by a significant decomposition (Moffat *et al*., 2005). Dapsone is widely employed as effective antibiotic for prophyl axis agent pneumocystis carinii pneumonia and an opportunistic disease in (HIV) infection. It's approved as antibiotic by food and drug adminstration since 1963. (Saillourglenisson *et al*., 2000). Dapsone is a medication most commonly used in combination with rifampicin and clofazimine as multidrug therapy (MDT) for the treatment of mycobacterium leprae infections (Leprosy), and with pyrimethamine in the treatment of malaria (Croft, 2007 ; Alkadi, 2007).

Among the various methods available for the estimation of dapsone are electrophoresis, gas chromatography and HPLC. Spectrophotometry is still a preferred technique due to its simplicity. Several spectrophotometric methods are available for the estimation of dapsone using diazotization and coupling with α-naphthol in the presence of sodium carbonate (Mohammed, 1994), dibenzoylmethane in an alkaline medium (Revanasiddappa and Manju, 2001), N-(1-naphthyl)ethylenediamine dihydrochloride (N-NED) in a hydrochloric acid medium (Nagaraja *et al*., 2001), iminodibenzyl in alcohol medium (Nagaraja *et al*., 2002), 3-aminophenol in aqueous medium (Nagaraja *et al*., 2003), sodium 1,2-naphthaquinone-4-sulfonic as the chromogenic reagent (Wang *et al*., 2004), benzoylacetone in alkaline medium (Omran, 2005), α-naphthol in strong alkaline medium in the presence of cetavlon (Al-Ramadani, 2007), phloroglucinol in basic medium (Daood, 2008).

Charge-transfer reactions are used for the determination of dapsone using different reagents such as chloranil (Mahmood, 2000), flouranil (Al-Gabsha *et al*., 2004), 2,3-dichloro-5,6-dicyano-benzoquinon (DDQ) (Al-Gabsha *et al*., 2004).

Other methods used oxidative coupling reaction with different reagents such as: promethazine in the presence of hypochlorite as oxidizing agent (Al-Abachi *et al*., 1995), 4-amino-N,N-diethylaniline in the presence of dichromate in acidic medium (Al-Abachi,

1997) by using the same method (Al- Talib,1997) replaced the reagent with 4-amino-N,N-dimethylaniline, and N,N-diethyl-p-phenylendiamine sulphate in the presence of KIO_4 was used (Nagaraja *et al*., 2010).

Liquid chromatography has been used for the determination of dapsone in human plasma (Shirazi *et al.*, 2001), in meat and milk (Hadjigeorgiou *et al*., 2009).

Other chromatographic methods have been used for the determination of dapsone in human plasma using HPLC technique (Queiroz *et al*., 1997), and HPLC with ultraviolet (Kwaddijk and Torano, 2002) and high-speed gradient liquid chromatography in serum (Luan Chen *et al*., 2003). Finally, the potentiation of dapsone induced methemoglobinemia by N-acetyl cysteine in rats (Moraes *et al*., 2008).

This paper describes simple and sensitive method. The first method included spectrophotometric determination of dapsone via oxidative coupling reaction with pyrocatechol in presence of potassium periodate in acidic medium. The second method included the developed HPLC method, by using C_{18} column with water: acetonitrile (50:50) as a mobile phase. The methods have been applied successfully to the determination of dapsone in pharmaceutical preparation (Tablet).

EXPERIMENTAL

Apparatus

Spectral and absorbance measurements are carried out using Shimadzu UV-Visible Recording Spectrophotometer UV-160, with 1-cm plastic cells. The pH measurements were carried out using HANNA Instrument 211 microprocess pH meter.

A Shimadzu LC-20 AD HPLC system with C18 stainless steel column (250 mm×4.6 mm) have been used in the analysis. Pump pressure 4.5-5.1 MPa and 20 µL is injected. The mobile phase consisted of (50: 50 V:V) water: acetonitrile. Ambient temperature, flow rate (0.5 ml /min.) and detector wavelength (297 nm).

Reagents

All chemicals used are of the pure analytical grade. Chemicals for HPLC are of analytical HPLC grades. Water used is double distilled and filtered using membrane filter.

Extraction of dapsone from tablets (British Pharmacopoeia, 2009).

10 tablets of dapsone were grinded and dissolved in acetone, the precipitate of dapsone was recrystalized (Ferry *et al*., 1964) twice to yield a pure dapsone of 178 ˚C as a melting point (175-181˚C).

Dapsone solution, 100 µg / ml.

This solution was prepared by dissolving 0.01 g of recrystallized dapsone mentioned above in 3 ml of ethanol and completed the volume to 100 ml with distilled water in a volumetric flask then the solution was kept in a brown bottle.

Pyrocatechol reagent solution.

Pyrocatechol reagent solution (2.7×10^{-3}M) was prepared by dissolving 0.03g of pyrocatechol (Fluka) in distilled water and the solution is diluted to 100 ml in a volumetric flask. The solution was transferred to a brown bottle and remained stable for only 24 hr.

Potassium periodate solution KIO_4, 0.1% .

This solution was prepared by dissolving 0.1 g of potassium periodate (Fluka) in 100 ml of distilled water in a calibrated flask. The solution is kept in a brown bottle and was stable for at least one week.

Hydrochloric acid solution, 0.1M .

This solution was prepared by diluting 10 ml of 1M hydrochloric acid to a 100 ml with distilled water in a calibrated flask.

Recommended procedure and calibration graph

Aliquots of dapsone solution were placed into 25-ml calibrated flasks so that their final amounts concentrations were between 10 to 500 µg. To each dapsone solution, 1.5 ml of 2.7×10^{-3}M pyrocatechol reagent solution, 2 ml of 0.1 M hydrochloric acid, 2 ml of 0.1 % KIO_4 and distilled water up to final volume of 25 ml were added. After 2 minutes, the absorbance of each coloured solution at 509 nm against a similarly prepared reagent blank was measured. The linear relationship between absorbance and concentration of dapsone was observed up to 500 µg, as shown in Fig. (1). The molar absorptivity has been found to be 1.05×10^{4}l .mol^{-1}.cm^{-1}, and Sandell sensitivity index 0.0235 µg. cm^{-2}. while the limit of detection (LOD) was found to be 0.1735 µg /ml and the limit of quantitation (LOQ) was found to be 0.5785 µg /ml (Valcared, 2000).

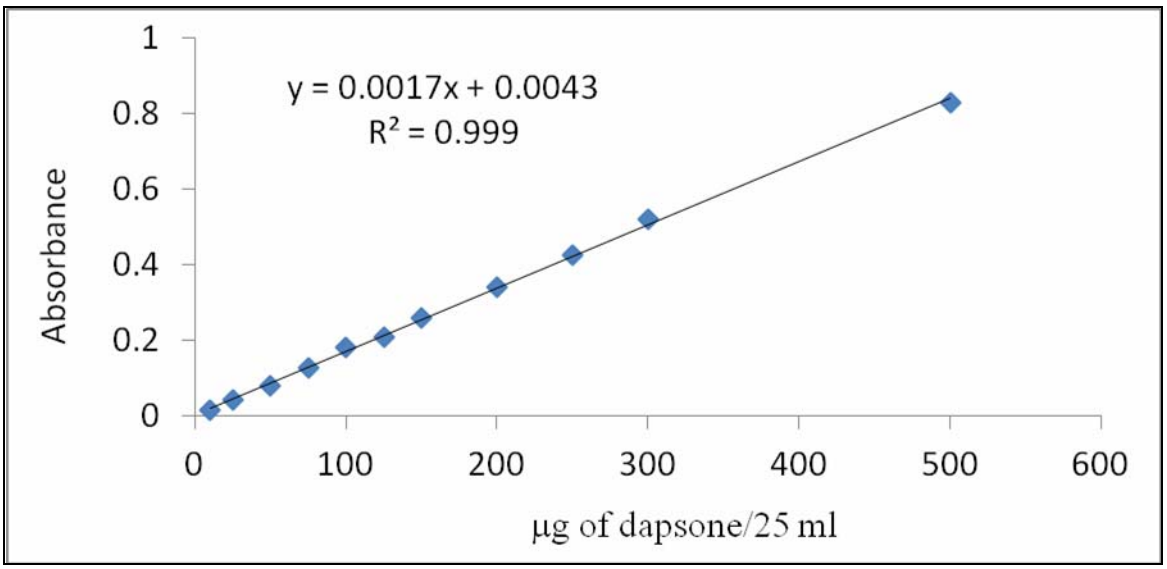


Fig. 1: Calibration graph for dapsone spectrophotometric determination.

HPLC method:

Dapsone standard solution was prepared in concentrations between 0.5-20 µg / ml in a mobile phase. The mobile phase consisted of (50:50) (v:v) water: acetonitrile isocratically eluted. Twenty µL of each standard solution was injected to C_{18} column, the mobile phase flow rate equal to 0.5 ml/min., the peak area of dapsone was plotted as a function of dapsone concentration at ambient temperature. The peak of dapsone was followed at 6.7 min.

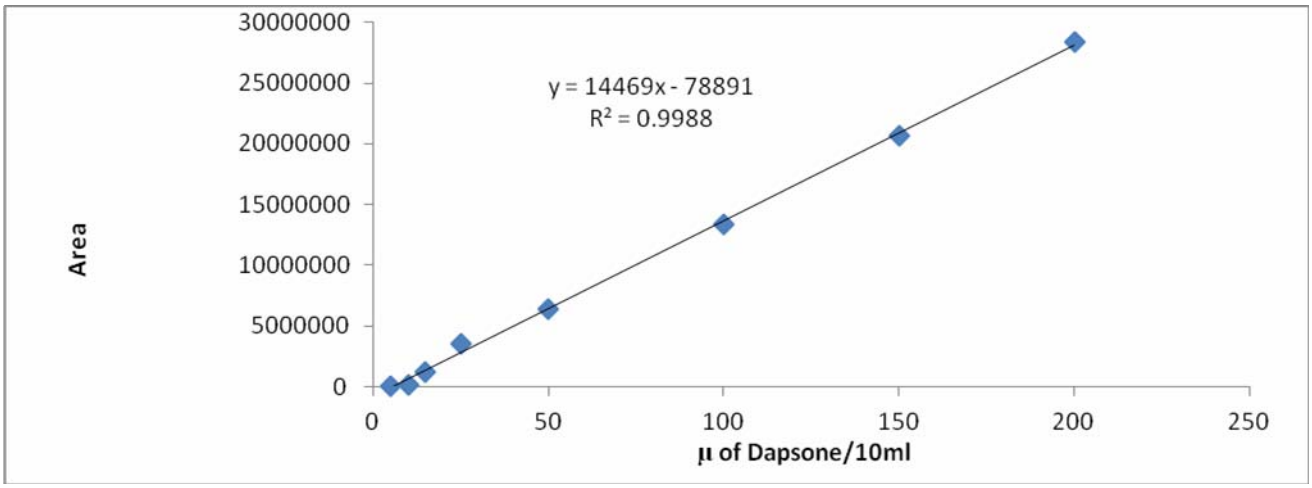


Fig. 2: Calibration graph of HPLC method.

A linear calibration graph is obtained between the area under the curve and the concentration over the range 0.5 -20 µg. ml^{-1} of dapsone was obtained.

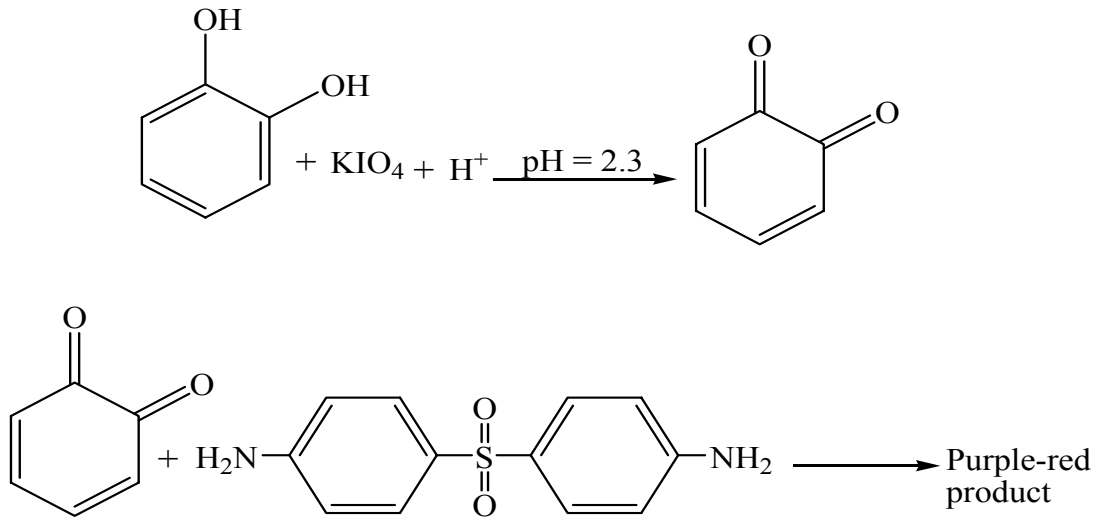
Procedure for dosage form:

For tablets, ten tablets (100 mg Dapsone / tablet) of the drug were weighed, powdered and mixed well. A portion equivalent to 0.01g was weighed and dissolved in 50 ml of distilled water, shaken well, filtered and diluted with water to 100 ml in a volumetric flask. An aliquot of diluted drug solution was then treated as done in the recommended procedure.

RESULTS AND DISCUSSION

Principle of the colour reaction:

Dapsone reacts with pyrocatechol in the presence of potassium periodate in acidic medium:



Study of the optimum reaction conditions

The various parameters which affect the colour intensity of the complex have been studied and the optimum conditions were selected.

Effect of oxidizing agents:

The effect of some general oxidizing agents on the intensity of the coloured dapsone – pyrocatechol dye was studied by adding 3 ml of pyrocatechol (2.7×10^{-3}M), 3ml of different oxidizing agents (0.1 %), potassium periodate, potassium dichromate, potassium chromate, potassium iodide, N-bromosuccinimide, N-chlorosuccinimide and ammonium ferric sulphate, 2 ml (100 $\mu g.ml^{-1}$) of dapsone and 2 ml (0.1M) hydrochloric acid solution. The volumes were completed with distilled water to the mark in a 25-ml volumetric flasks. The absorbance was measured at 515 nm. From the experimental data, it can be stated that potassium periodate gave good results with a low blank absorbance value, which was selected for subsequent experiments.

Effect of acids:

Various acids (hydrochloric acid, nitric acid, sulphuric acid, phosphoric acid, acetic acid and formic acid) with different amounts had been investigated to examine their effect on the intensity of the coloured dye. The results revealed that the 1 ml of 0.1 M hydrochloric acid solution gave the best results.

Effect of pH:

The influence of pH on the colour intensity of the complex formed has been studied by transferring 3 ml of 2.7×10^{-3}M solution of pyrocatechol reagent, 2 ml of 0.1 % potassium periodate solution, 200 µg of dapsone and different volumes of 0.1M HCl and 0.1M NaOH, separately. The volumes were then completed with distilled water to 25 ml in volumetric flask. The absorbance were measured at 515 nm for each solution and the final pH was measured.

From the experimental data, the higher absorbance was observed to occur in acidic medium at pH 2.1 – 2.7. Further, colour formed is stable and contrasted. In basic medium, the coloured was different and the sensitivity had been lowered and a ccompanied by a weak contrast. So, the acidic medium had been selected for the subsequente experiments.

Effect of dapsone amounts to the reagent amounts:

To study this effect, different volumes of pyrocatechol (2.7×10^{-3}M), 2 ml of 0.1% potassium periodate, various amounts of dapsone (10, 25, 50,75,100, 125, 150, 200 $\mu g. ml^{-1}$) and 2 ml of hydrochloric acid solution (0.1M). The solutions were mixed and diluted with distilled water to the mark into a 25 ml volumetric flask, and then the absorbance was measured at 515 nm against a reagent blank prepared in the same manner.

The results summarized that 1.5 ml of (2.7×10^{-3}M) pyrocatechol and 200 $\mu g.ml^{-1}$ of dapsone gave the best absorbance and the correlation coefficient is 0.998 which gave the maximum colour intensity. The optimum values are selected in all subsequent experiments.

Effect of temperature:

The effect of temperature on the colour intensity of the coloured product was studied. This was performed by placing into three 25-ml calibrated flasks, the first one is allowed to stand for increasing time intervals at room temperature, the second at 0 ºC, and the third at 60 ºC using water bath.

The results recommended that the absorbance of the colour purple-red was decreased when the reaction was carried out at 0 ºC or 60 ºC. Therefore, the reaction mixture should be carried out at room temperature.

Effect of delay time:

The coloured product was developed rapidly after the addition of the oxidant and exhibited a maximum intensity at room temperature, different delay times was examined the data showed that the development time was 2 minutes which was allowed before measuring the samples.

Effect of surfactants:

The effect of the presence of surfactants on the colour intensity of dapson-catechol dye was examined, cetyl pyridinium chloride (CPC) and cetyltrimethyl ammonium bromide (CTAB) (cationic surfactant), sodium dodecyl chloride (SDS) (anionic surfactant) and Tween 80 (nonionic surfactant) were tested. The results reveal that the presence of surfactants have no beneficial effect at all. Therefore, it has been recommended to eliminate the use of surfactants in the subsequent experiments.

Order of addition:

From the different possible orders of addition of reagents, the order (Dapsone+Reagent+HCl+KIO_4) is selected for the subsequent experiments, otherwise a loss in colour intensity is observed.

Effect of time:

To determine the suitable time for absorbance measurement, the effect of time on the development and stability of the coloured complex was investigated under optimum conditions at the wavelength of maximum absorption at 509 nm. Table (1) showed the result that the stability period of at least 60 min is sufficient for many measurements to be made.

Table 1: Effect of Time.

Time (min)	Absorbance / µg of dapsone present				
	10	50	100	200	400
ADTTM*	0.008	0.066	0.128	0.346	0.624
5	0.006	0.066	0.128	0.346	0.626
10	0.011	0.066	0.127	0.344	0.626
15	0.010	0.067	0.126	0.344	0.624
20	0.010	0.061	0.127	0.342	0.622
25	0.010	0.063	0.126	0.342	0.622
30	0.014	0.061	0.124	0.341	0.622
35	0.014	0.061	0.124	0.342	0.620
40	0.013	0.063	0.121	0.340	0.620
45	0.007	0.059	0.118	0.340	0.620
50	0.006	0.057	0.119	0.340	0.621
55	0.004	0.052	0.115	0.340	0.619
60	0.004	0.052	0.115	0.339	0.619

*ADTTM : After dilution to the mark

Final absorption spectra:

When dapsone and pyrocatechole reagent solution are mixed in the presence of KIO_4 in acidic media, an immediate purple-red coloured complexes are formed. Fig. (3) shows a single maximum absorption at 509 nm, whereas the reagent blank gives a maximum absorption at 389 nm.

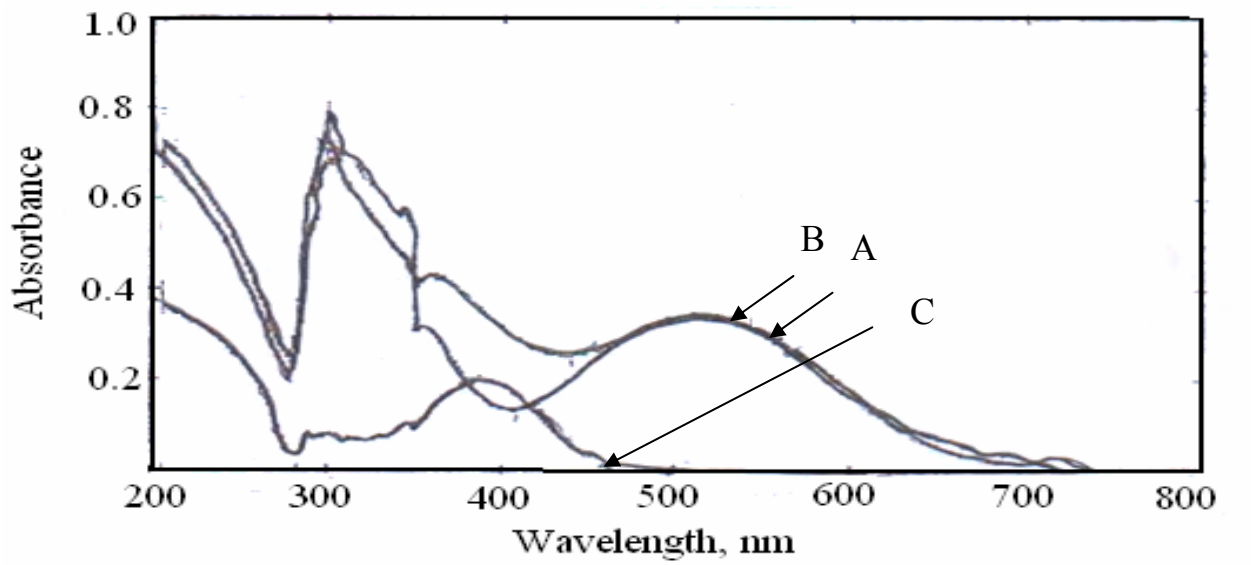


Fig. 3: Absorption spectra of 200 µg / 25 ml measured, (A) against blank, (B) against distilled water, (C) blank against distilled water

The 509 nm wavelength of maximum absorption is still being adapted for subsequent experiments.

Accuracy and precision:

The accuracy and precision had been checked by determining dapsone at four different concentrations (25, 50, 100, 200) µg. The results showed that the recoveries were between (100 to101.1)% and the relative standard deviations were ranged between ± 2.60 to ± 1.05, indicating a satisfactory method.

Nature of the dye:

The composition of the intense purple-red dye has been established using Job's method of continuous variations. Fig.(4) has shown that the dapsone and pyrocatechol reagent combine in a 1:2 ratio.

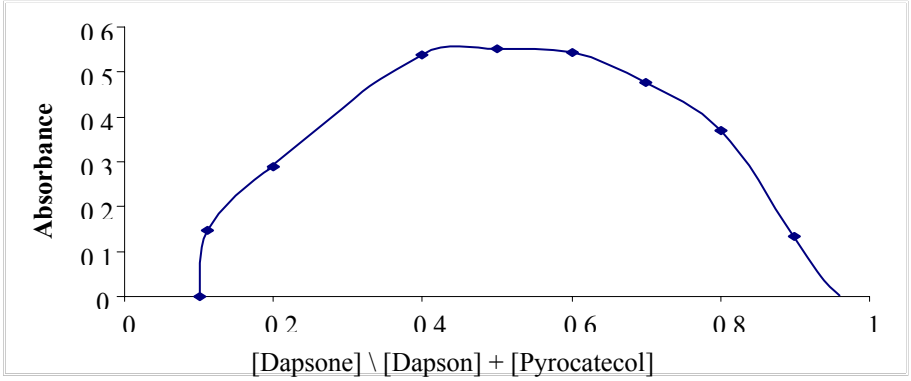
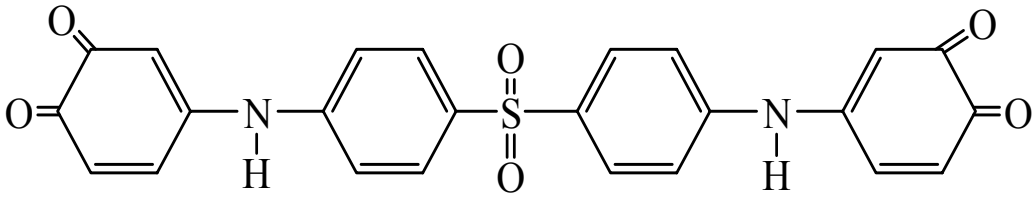


Fig. 4: Continuous variations plot for dapsone –pyrocatechol coloured product

Therefore, the structure of the formed coloured product might be represented as follows:



Purple-red product

Effect of organic solvents:

The spectral characteristics of the purple-red coloured product in various organic solvents are shown Table (2). Water is shown to be a good medium from the point of view of sensitivity and economy, therefore it has still been recommended for dilution.

Table 2: Spectrophotometric characterization of the coloured product in various Organic solvents.

Solvent	λmax, nm	ε, $l.mol^{-1}.cm^{-1}$
Acetic acid	512	5.8×10^4
Acetone	494	4.9×10^4
DMF	478	4.6×10^4
Ethanol	480	2.8×10^4
Formic acid	546	4.7×10^4
Methanol	-----	Turbid
Propanol	494	5.0×10^4
Pyridine	560	2.5×10^4
Water	510	6.3×10^4

Study of interferences:

In order to assess the possible analytical application of the present proposed method, the effect of foreign compounds had been studied by carrying out the determination of 200 µg of dapsone in the presence of different amounts (50, 100, 200, 400) µg of each interferent using the recommended procedure.[Table (3)].

Experimental results showed that there was no interference from excipients for the examined method.

Table 3: Effect of interferences.

Interferences	Recovery, % of 200 µg of dapsone per µg interference			
	50	100	200	400
Acacia	97.4	100.3	95.9	95.9
Dextrose	102.3	99.1	98.8	98.5
EDTA	99.3	99.3	98.4	98
Glucose	100.3	100.5	100.8	100.9
Glycerol	98.6	99.3	99.3	99.7
Lactose	98.1	97.3	100.3	100.8
Menthol	100.5	100.3	100.3	100.2
Starch	100.3	101.7	102.6	102.5
Sucrose	101.7	101.4	100.5	100.2

Application of the method:

The developed method had been applied to the determination of dapsone in pharmaceutical preparation (Tablet), the results are shown in Table (4).

Table 4: Determination of Dapsone in tablet.

pharmaceutical preparation	Certified value (mg) tablets	µg dapsone present / 25 ml	µg dapsone found / 25 ml	Recovery*, (%)
Dapsone	100	50	51.7	103.4
		100	100.6	100.6
		200	201.2	100.6
		300	300	100

- Average of five determinations.

For comparison, standard addition method (Skoog *et al*., 2000) was used for the determination of dapsone under investigation in order to prove that the proposed method is applied in the determination of dapsone without interferences [Table (5) and Fig.(5)].

Table 5: Determination of dapsone using standard addition method.

pharmaceutical preparation	Certified value (mg) tablets	µg dapsone present / 25 ml	µg dapsone found / 25 ml	Recovery*, (%)
Dapsone	100	200	203.10	101.5
		300	301.06	100.3

* Average of four determinations.

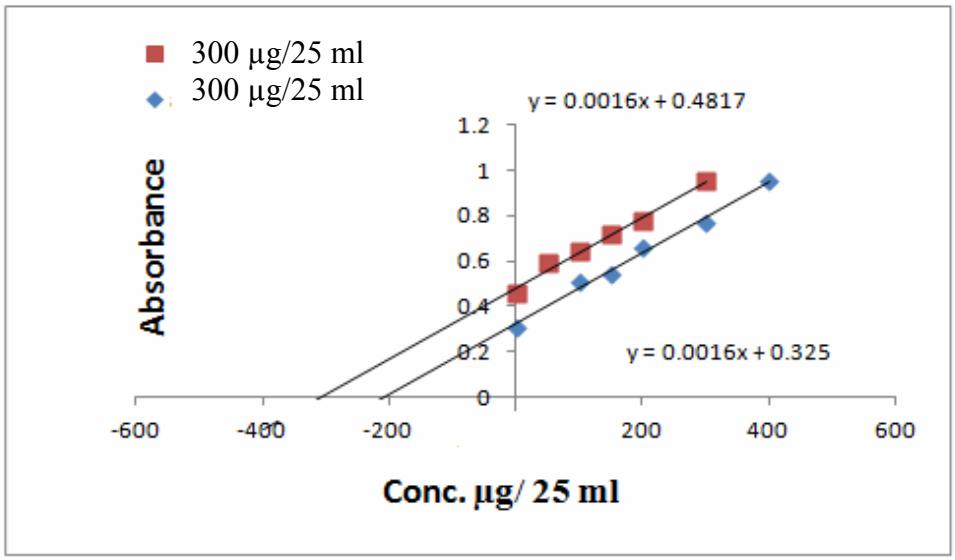


Fig. 5: Graph of standard addition method for the determination of dapsone in tablets.

The results in Table (5) and Fig. (5) indicating that the suggested method for the determination of dapsone can be used with satisfactory results.

Both the present and standard addition methods have been applied at the same time to F-test and t-test calculation and the value compared with the statistical tables for six degrees of freedom at 95% confidence level. The results in Table (6) showed that there is no real difference between the two methods.

Table 6: The results of F and t-test analysis

Drug	**Pharmaceutical preparation**	**Recovery *%**		**F-test**	**t-exp.**
		Present method	**Standard addition**		
Dapsone	Tablet	100.5	101.5	9.217	0.69

- Average of four determinations.

Comparison of the method:

Table 7 is showing a comparison between some analytical variable for the present method and those of the recent spectrophotometric methods.

Table 7 : Comparison of the methods

Analytical parameters	Present method	Literature method (Al-Abachi, 1995)	Literature method (Al-Talib, 1997)	Literature method (Nagaraja, 2010)
Reagent	Pyrocatechol	Promethazine hypochlorite	4-amino-N,N-diethylaniline	N,N-diethyl-p-phenylenediamine sulphate
λmax (nm)	509	604	551	550
Colour of dye	Purple-red	Bluish-green	Red-violet	Red
Beer's law (ppm)	0.4-20	0.2-4	0.1-36	1.5-12
Molar absorptivity ($l.mol^{-1}.cm^{-1}$)	1.05×10^4	2.9×10^4	1.34×10^4	1.66×10^4
pH	2.4	2.5	2.8	-----
Medium	Aqueous	Aqueous	Aqueous	Organic
Temperature, C°	R.T	R.T	R.T	60±5
Recovery, %	100-101.1	100.26-101.6	-----	99.5-99.9
RSD, (%)	1.05-2.60	0.27-0.54	Less than 2%	0.1-2.2
LOD (mg/ml)	0.0145	-----	-----	0.44
Analytical application	Tablets	Tablets	Tablets	Pharmaceutical dosage forms

HPLC method

Selection of wavelength

The absorption spectrum of 25 µg $.ml^{-1}$of dapsone in 50:50 (v:v) water: acetonitrile has been taken, Fig. (6) shows that maximum absorbance of dapsone is at 297 nm. Therefore, 297 nm has been used for UV-detection.

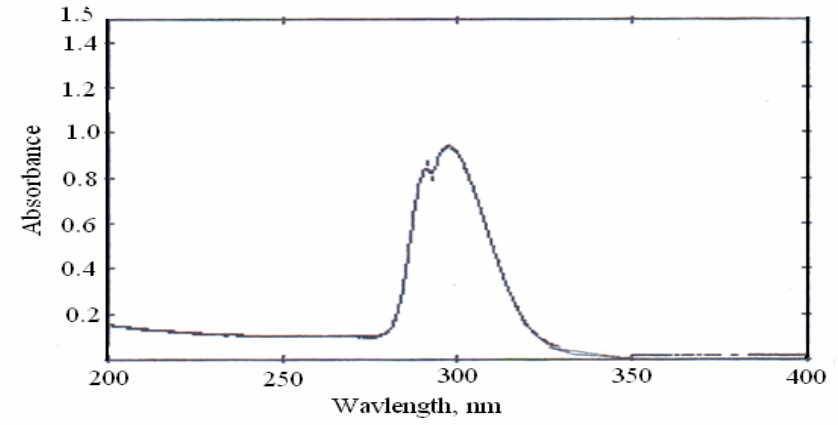


Fig. 6: Selection of wave length.

Selection of mobile phase

Different polar solvents with various compositions (used as a mobile phase) have been used to construct the optimum composition. The retention time and capacity factor have been followed in Table (8) and Fig. (7-1).

Table 8: Selection of the mobile phase

Mobile phase composition	Retention time, min.	Capacity factor, K
Water: methanol (60:40)	5.36	1.329
Water: acetonitrile (60:40)	5.72	1.690
Water: acetonitrile : methanol (75:20:5)	11.63	4.631

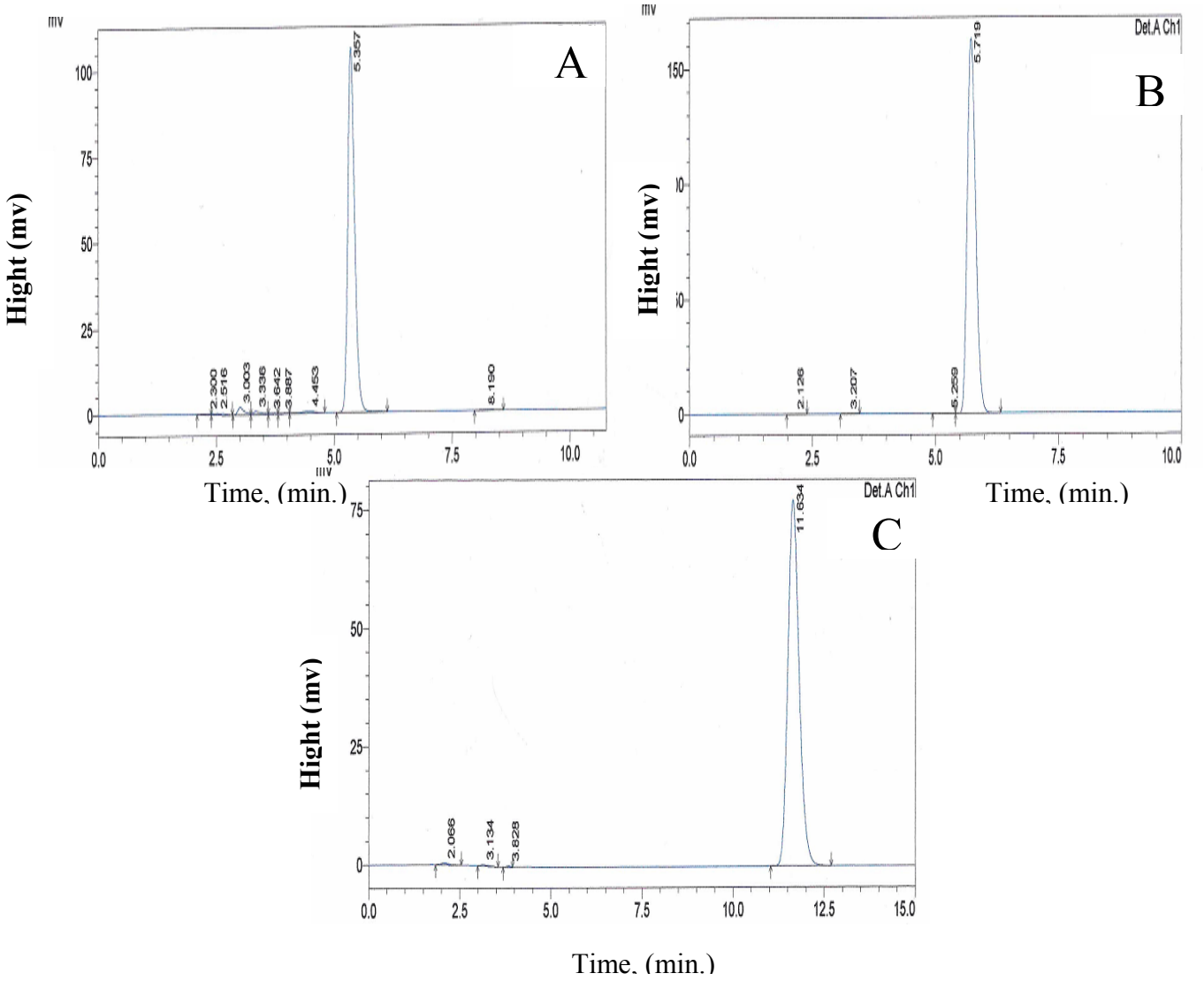


Fig. (7-I): Selection of mobile phase.
A = Water: Methanol (60:40)
B = Water: Acetonitrile (60:40)
C = Water: Acetonitrile: Methanol (73:20:5)

Fig. (7-I) shows that the case B is more useful because the resolution is better than the others, so that various volumes ratio of water: acetonitrile studied as mobile phase Fig.(7-II) shows that the graph is more useful because the resolution is the best and it is associated with higher sensitivity. Therefore, the condition reported in Fig.(7-II) C is recommended.

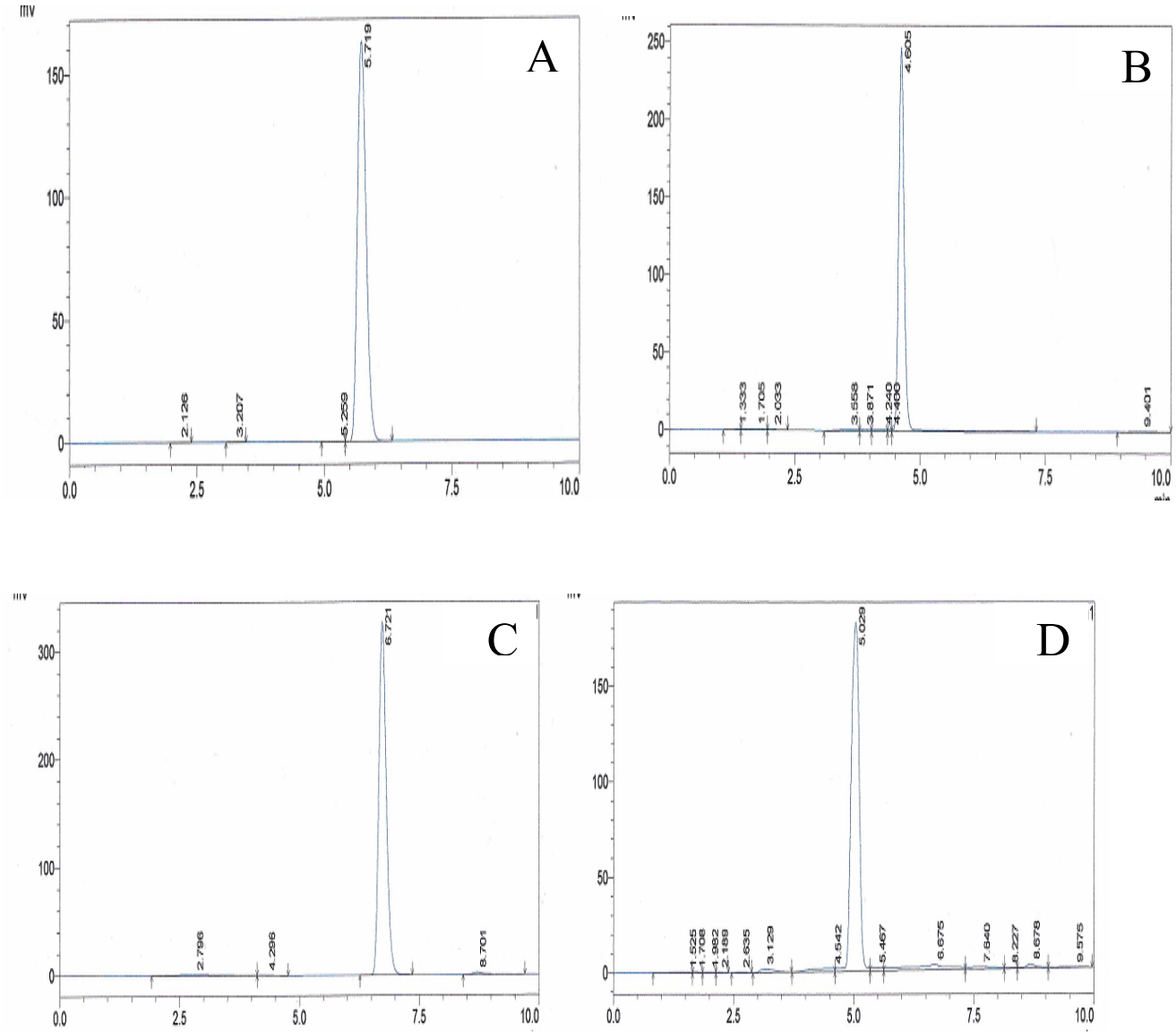


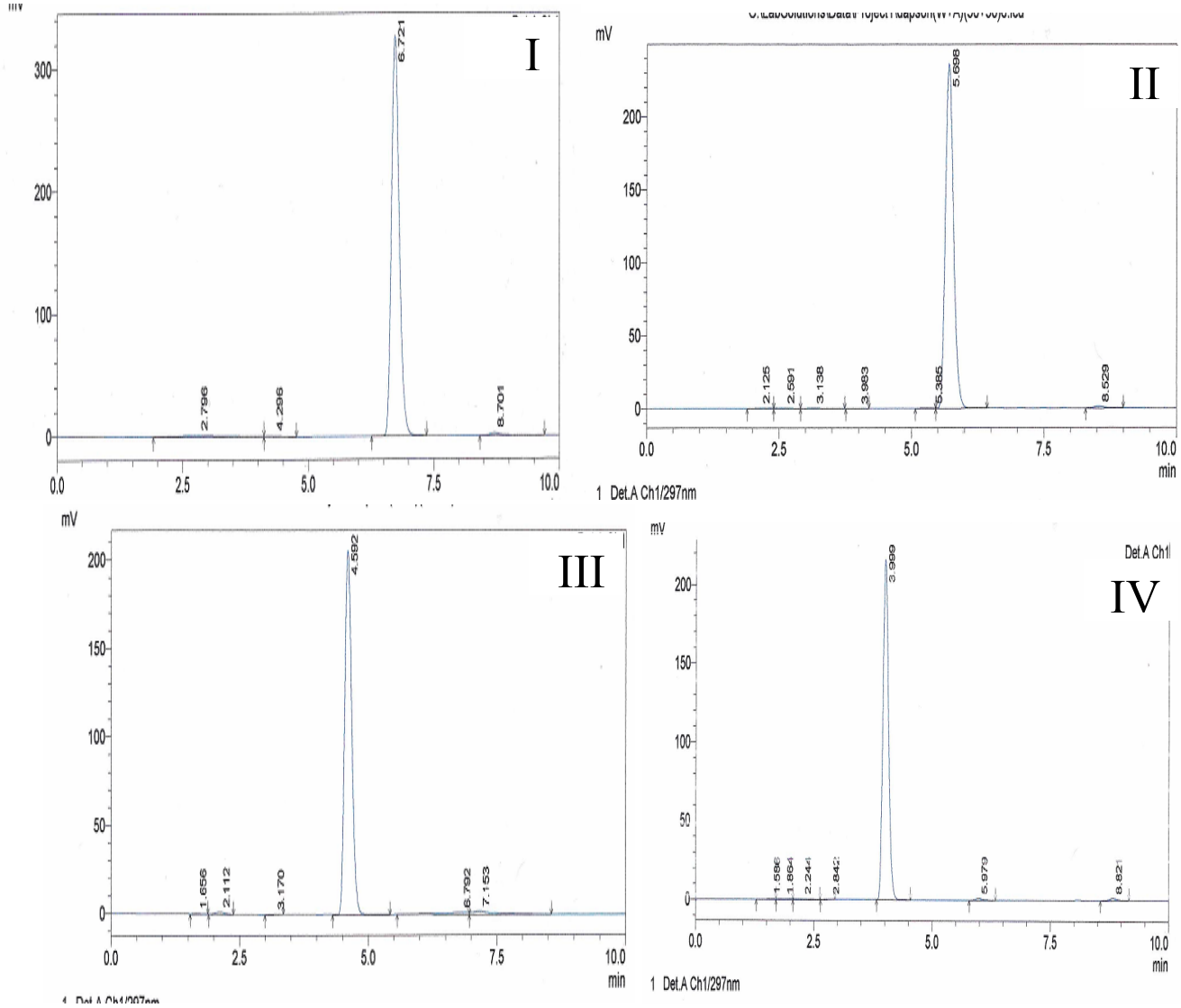
Fig. (7-II): Selection of volumes ratio of mobile phase.
Water : Acetonitrile
A = (60:40)
B = (70:30)
C = (50:50)
D = (40:60)

Selection of flow rate:

A flow rate (0.5-1.2) ml.min^{-1} as a flow rate has been studied, 0.5 ml.min^{-1} gives the optimum capacity factor Table (7) with clear chromatography and good sharpness Fig. (8).

Table 9: Selection of floe rate

No.	Flow rate (ml/min.)	Retention time (min.)	Capacity factor K
I	0.5	6.721	1.40
II	0.7	5.698	1.68
III	0.9	4.592	1.77
IV	1.0	3.999	1.52
V	1.2	3.365	1.63



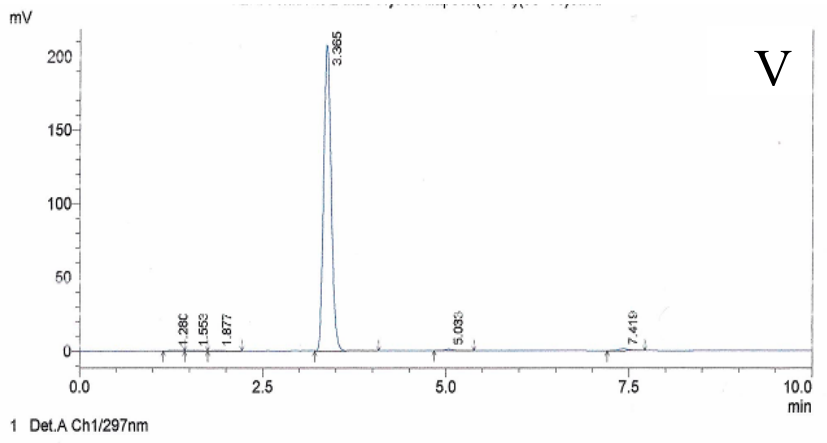


Fig. 8: Selection of flow rate

Accuracy and precision

To study the accuracy and precision of the calibration graph, dapsone was determined at three different concentrations and the results are shown in Table (10), which indicate a good accuracy and precision.

Table 10: Accuracy and precision.

Amount of dapsone µg, taken	Relative error, %*	Relative standard deviation, %*
10	0.3	± 0.83
25	0.0	± 0.53
50	1.1	± 1.3

* Average of three determinations.

CONCLUSIONS

The suggested methods for the determination of dapsone are sensitive and temperature independent, applicable without resorting to an extraction step.

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