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Preparation and Characterization of Spironolactone-Avicel PH 101 physical mixtures and adsorbates.

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ABSTRACT

The objective of this study was to investigate the effect of the surface adsorption of poorly soluble drug spironolactone onto an inert matrice Avicel PH 101 as a technique for improving the drug dissolution. The loaded mixtures of spironolactone with Avicel PH 101 were prepared by solvent deposition method. Adsorbate systems containing (1:1, 1:2 and 1:3) w/w drug/adsorbent ratios were prepared. The adsorbates were scrapped and powdered. Also physical mixtures of different w/w drug/carrier ratios (1:1, 1:2 and 1:3) were prepared by simple mixing. The physicochemical characterization of the systems using differential scanning calorimetry (DSC) and powder X-ray diffraction was carried out to detect possible interaction between the drug and the carrier, moreover, quantitative solubility and in-vitro dissolution studies of spironolactone alone and in physical mixtures or adsorbates were studied in simulated gastric fluid (SGF) of PH 1.2 and in simulated intestinal fluid (SIF) of PH 7.4.

The reduction of drug peaks in DSC profile of the adsorbate particles suggests the transformation of crystalline spironolactone into an amorphous from. The same result is indicated by X-ray diffraction, where the adsorbate mixture showed smaller characteristic peaks than those of the plain drug with reduced intensity indicating incomplete transformation of the drug to the amorphous state. The study showed a little improvement in the dissolution pattern of the drug in case of the adsorbate and a more improvement in case of physical mixtures.

INTRODUCTION

Spironolactone is a steroidal drug acting as a specific aldosterone antagonist used as potassium sparing diuretic. It is described as a light, cream-colored to light tan, crystalline powder with a faint to mild mercaptan-like odor, practically in soluble in water, soluble in alcohol and in ethyl acetate, freely soluble in chloroform and in benzene, and slightly soluble in methyl alcohol and fixed oils (USP XXIX, 2006). It is a poorly soluble drug substance, which has been reported to have difference in solubility (Agafonov *et al.*, 1991). Because of its low water solubility and slow dissolution rate, spironolactone shows a variable and incomplete oral absorption (Seo *et al.*, 1993).

Several techniques have been developed to optimize the dissolution rate of poorly water soluble drugs, like spironolactone, to improve their bioavailability. Such methods include particle size reduction, solubilization, salt formation and preparation of solid dispersion systems (Kaukonen *et al.*, 1997, Soliman *et al.*, 1997, Serajuddin, 1999, Liversidge *et al.*, 2003, Stegeman *et al.*, 2007, Yassin *et al.*, 2009).

The objective of this work was to prepare physical mixtures and adsorbate mixtures of spironolactone with Avicel PH 101 and investigate the effect of this technique as a way to improve the solubility and dissolution of the drug.

Materials and Methods

Preparation of the adsorbate

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The calculated amount of the adsorbent Avicel PH 101 was added to the chloroformic solution of spironolactone to give the desired drug/absorbent ratios. Adsorbate systems containing (1:1, 1:2 and 1:3) w/w drug/adsorbent ratios were prepared. Chloroform was evaporated at room temperature. The samples were then scrapped and powdered, then assayed for their drug content.

Preparation of physical mixtures

The physical mixtures of the drug and the adsorbent in different drug/adsorbent ratios (1:1, 1:2 and 1:3) were prepared by simple mixing avoiding any grinding effect.

Characterization of the prepared physical and adsorbate mixtures

Determination of the drug content

An accurate weighed sample of the adsorbate equivalent to 7.5 mg of the drug was dissolved in simulated gastric fluid (SGF) using a sonicator. The volume was completed to 500 ml with SGF and the concentration of spironolactone was determined spectrophotometrically at 238 nm.

Differential scanning calorimetry

DSC was performed using DSC-60 Shimadzu instrument under the following conditions; sample weight range of 1-4 mg, heating rate of 10°/min, N_2 purge rate of 20 ml/min using aluminum pan hermetically sealed. The instrument was calibrated for temperature and energy with pure indium (99.999%, melting point 156.6° and transition energy 28.45 J/g). Thermal analysis was carried out using TA 60 PC system with Shimadzu software program.

X-ray diffractometry

The X-ray diffraction patterns of the selected samples were obtained using a computer Philips operating in two modes using copper tube radiation. A Cu target tube operated at a voltage of 40 KV and a current of 30 mA and a single crystal graphite monochromator were employed. An attached microprocessor utilizes a special software program to analyze peak position and intensities. Standard polycrystalline silicone was used to calibrate the equipment.

Quantitative solubility

Quantitative solubility of spironolactone, its physical mixtures and adsorbate mixtures was determined in SGF (pH 1.2) and SIF (pH 7.5) by the equilibrium solubility method, which employs a saturated solution of the material in the solvent for a prolonged period until equilibrium is achieved.

Preparation of saturated solution of spironolactone

For the determination of solubility, excess of spironolactone, its physical mixtures and its adsorbate mixtures was placed in contact with 30 ml of solvent (SGF pH 1.2 or SIF pH 7.5) in conical flasks, then covered with aluminum foil. The flasks were shaken 100 rpm for 5 hours at 37° C. It was found that equilibrium solubility was achieved by this period. After filtration, the concentration of drug in the saturated solution was determined spectrophotometrically at 238 nm after appropriate dilution with the solvent used. Each experiment was preformed three times and the mean was calculated in each case.

In-Vitro dissolution studies

The dissolution of the drug alone, from its physical mixtures and from its adsorbate mixtures was carried out using USP paddle dissolution apparatus. The dissolution medium was 900 ml SGF pH 1.2 or SIF pH 7.5, maintained at 37± 0.5° C. The paddles were positioned 2.5 cm from the bottom of the vessel and rotated at a speed of 75 rpm. For the test, accurately weighed amount equivalent

to 25 mg of the drug and prefilled in a hard gelatin capsule was transferred to the stirred dissolution medium in each vessel. At time intervals, 5 ml samples were withdrawn by a pipette fitted with a cotton plug at its terminal end. Equal volume of fresh dissolution medium was added to the dissolution medium so as to keep the volume of the dissolution medium constant. The absorbance of the samples was measured spectrophotometrically at 238 nm. Each experiment was performed three times and the mean was calculated in each case. The standard curve of spironolactone in SGF or SIF (according to the fluid used in dissolution) was used to determine the amount of drug dissolved. The cumulative amount of drug was calculated to compensate for the previously withdrawn samples.

RESULTS and DISCUSSION

Determination of the drug content

Table (1): shows the results of drug content determination. The drug content was 99.5, 102.5 and 110.7% for 1:1, 1:2 and 1:3 w/w (drug: adsorbent) adsorbate mixtures, respectively.

Table (1). Drug content for spironolactone-Avicel PH 101, adsorbate mixtures.

Drug: Adsorbent ratio	Theoretical drug content (mg)	Actual drug content (mg)	Drug content %
1:1	7.5	7.465	99.5
1:2	7.5	7.690	102.5
1:3	7.5	8.305	110.7

Differential scanning calorimetry

DSC analysis of plain drug, adsorbent, physical mixture and adsorbate mixture is shown in Figure (1). The DSC chart of plain drug exhibited a sharp endothermic peak at 206.29°. The DSC chart of adsorbent exhibited a shallow endothermic peak at 94.58° and a very little endothermic peak at 175.5°. The DSC chart of the physical mixture exhibited three endothermic peaks with little heights at 204.32, 236.19 and 245.39°. The DSC chart of the adsorbate mixture exhibited an endothermic peak with a little height at 189.06°. In case of the adsorbate, the melting endothermic peak of the drug was greatly reduced and appears with a little shifting, that may be attributed to transformation of drug to amorphous state.

X-ray diffractometry

X-ray diffractometry technique was utilized to study the crystallographic nature of plain drug, adsorbent, physical mixture and adsorbate mixture.

The major X-ray diffraction peaks of spironolactone were observed at 2 θ of 9.03, 15.85, 16.47, 17.08 and 20.16 with relative intensities of 71, 73, 92, 100 and 75, respectively. Chart D (figure (2)) of the adsorbate mixture showed smaller characteristic peaks than those of the plain drug with reduced intensity indicating incomplete transformation of the drug to the amorphous state.

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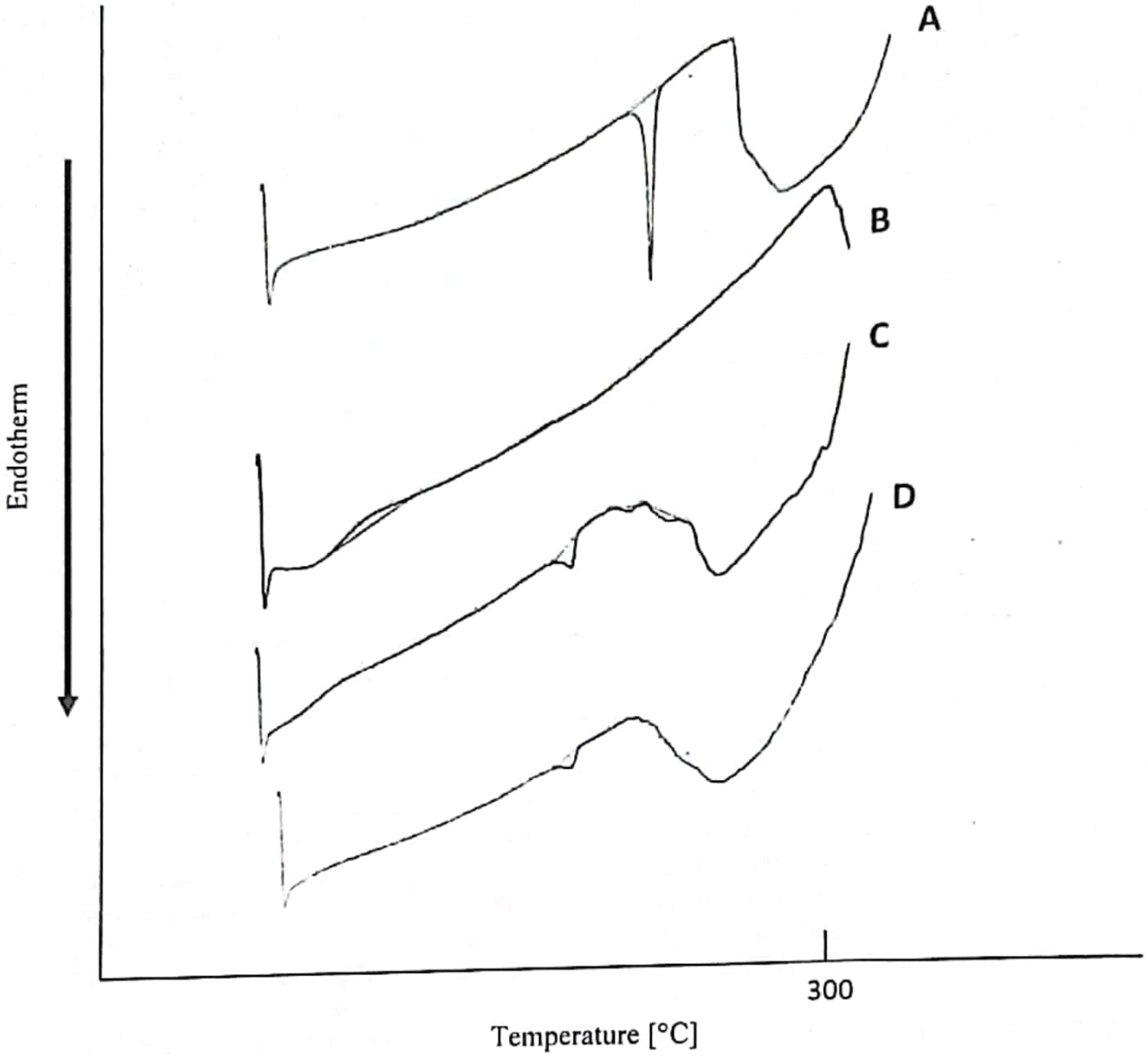

Figure 1. DSC thermograms of A- Plain drug, B- Adsorbent, C- Physical mixture and D- Adsorbate mixture.

Quantitative solubility

The solubility values of spironolactone, its physical mixtures and its adsorbates were calculated using the equilibrium solubility method. As it is shown in table (2) and figure (3), the solubility of plain drug was 39.36 µg/ml in SGF and 29.68 µg/ml in SIF. In case of physical mixture the solubility was 46.34, 39.32 and 38.96 µg/ml for 1:1, 1:2 and 1:3 drug/adsorbent ratios, respectively in SGF, where it was 31.47, 34.33 and 36.19 µg/ml for 1:1, 1:2 and 1:3 drug/adsorbent ratios, respectively in SIF. In case of adsorbate mixtures, the solubility was 35.21, 36.55 and 38.89 µg/ml for 1:1, 1:2 and 1:3 drug/adsorbent ratios, respectively in SGF, where it was 30.39, 34.33 and 38.91 µg/ml for 1:1, 1:2 and 1:3 drug/adsorbent ratios, respectively in SIF. The increased solubility of the drug in case of the adsorbate mixture

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may be attributed to the incomplete transformation of drug from the crystalline state to the amorphous state. The effect of the adsorbent for increasing the solubility of the drug is shown in case of SIF that may be due to the higher solubility of the adsorbent in the alkaline medium than that in the acidic medium (Wheatley, 2000).

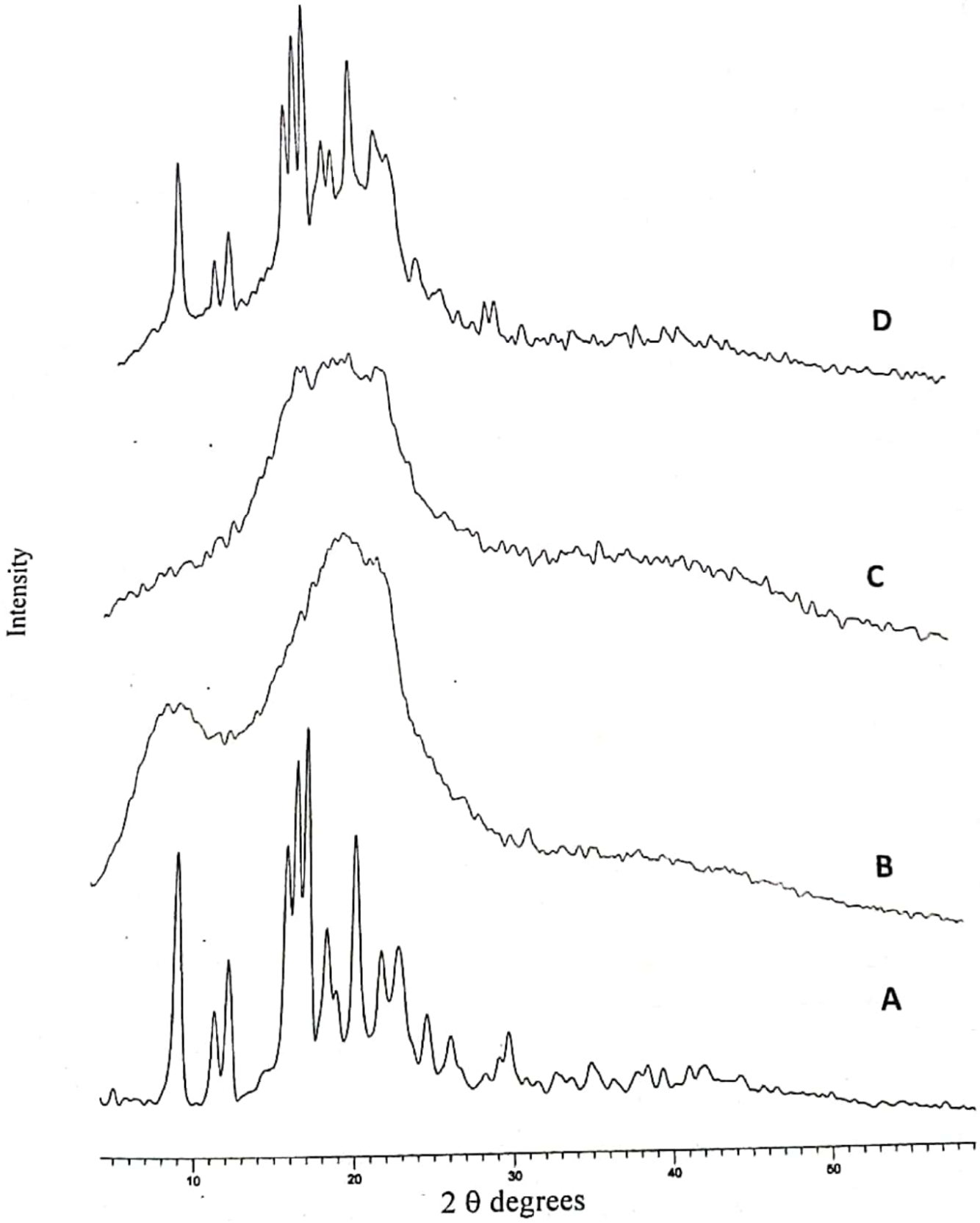

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Figure 2. X-ray diffraction patterns of A-Plain drug, B-Adsorbent, C-Physical mixture and D- Adsorbnte mixture

Table (2): Solubility of spironolactone, its physical mixtures and its adsorbate mixtures in SGF and SIF at 37° C.

Medium	Solubility µg/ml					
	Plain drug	Physical mixture			Adsorbate mixture		
		1:1	1:2	1:3	1:1	1:2	1:3
SGF	39.36	46.34	39.32	38.96	35.21	36.55	38.89
SIF	29.68	31.47	34.33	36.19	30.39	34.33	38.91

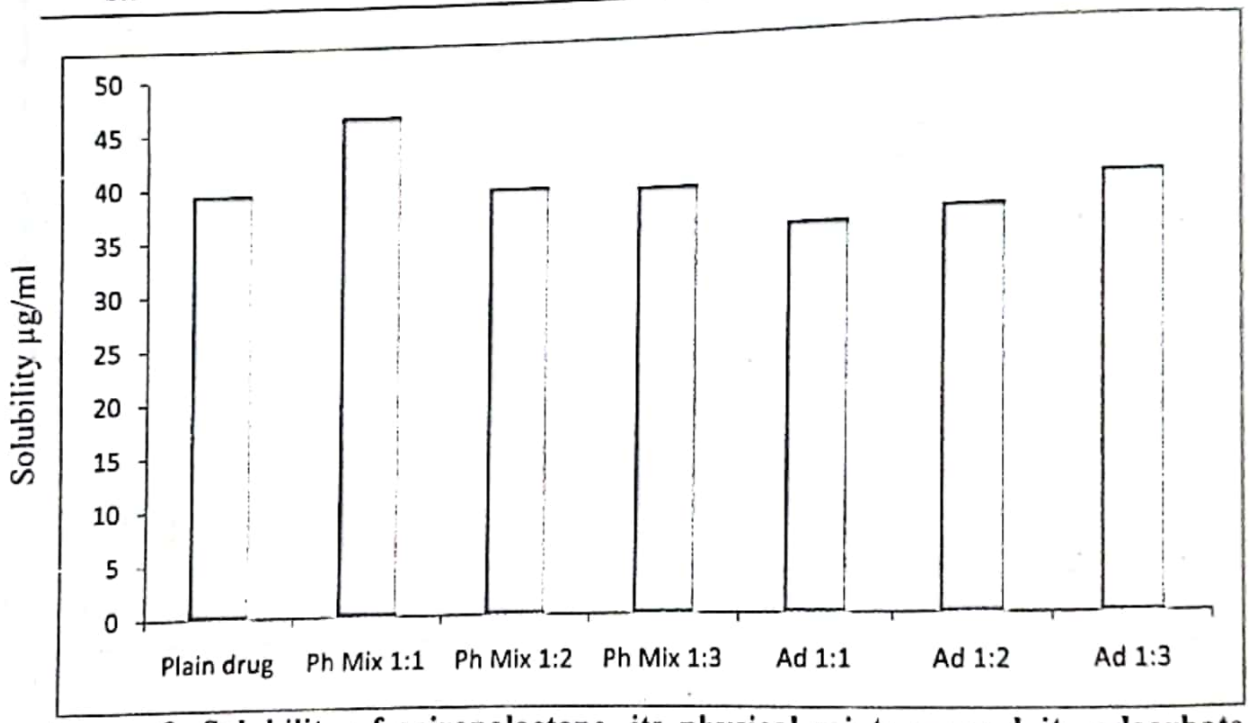

Figure 3. Solubility of spironolactone, its physical mixtures and its adsorbate mixtures in SGF pH 1.2 at 37° C.

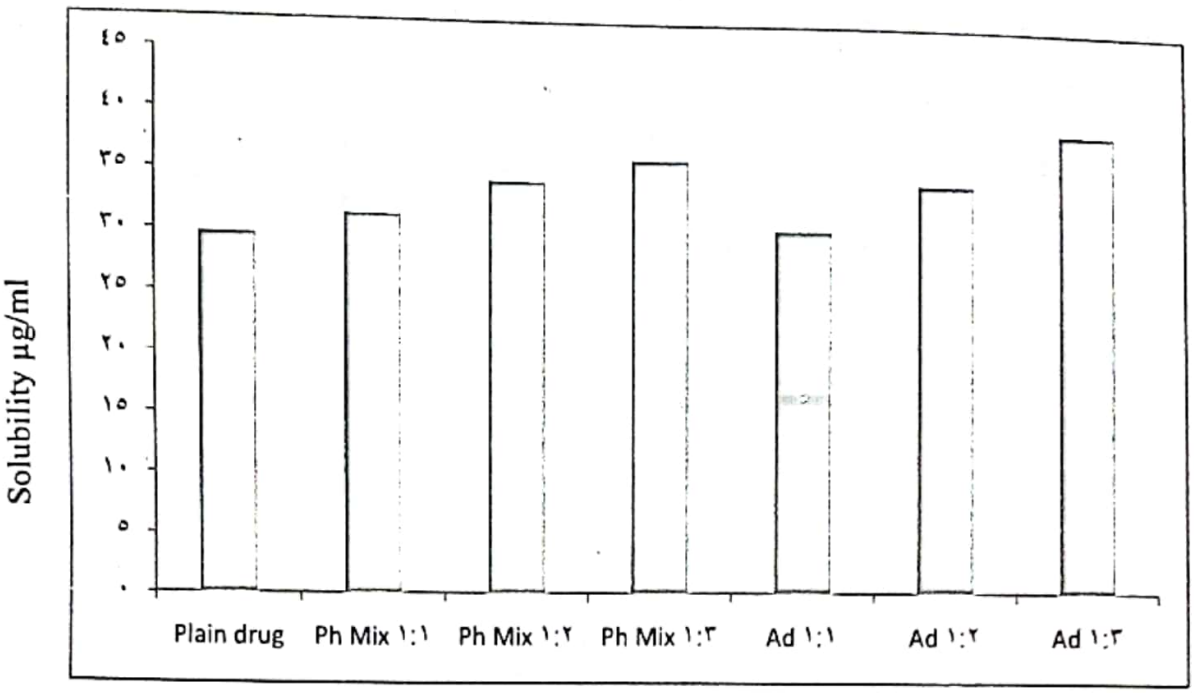

Figure 4. Solubility of spironolactone, its physical mixtures and its adsorbate mixtures in SIF pH 7.5 at 37° C.

***In Vitro* dissolution studies**

The results of the dissolution of spironolactone, its physical mixtures and its adsorbate mixtures with Avicel PH 101 are presented in tables (3, 4) and graphically illustrated in figures (5, 6). The results showed that the dissolution was increased upon using the physical mixtures of the drug in both of acidic and basic medium. The increased dissolution rate of spironolactone from physical mixtures may be due to the deaggregation of spironolactone clumps achieved by the carriers in the dissolution medium (Ismail *et al*; 2004). Tables (3, 4) and figures (5, 6) showed increase in dissolution of the drug upon using the adsorbate mixture in the ratio 1:3 drug to adsorbent. The percent released of spironolactone was higher in case of adsorbate mixture 1:3 drug to adsorbent ratio than the plain drug in both of SGF and SIF. The dissolution mechanism is therefore different from that of plain drug where dissolution in case of adsorbate mixture occurred due to rapid desorption of the physically adsorbed drug molecules, but in case of plain drug dissolution occurred from the surface of drug crystals according to the concentration gradient (Ali, 1997).

Table (3). *In Vitro* release of spironolactone, its physical mixtures and its adsorbate mixtures with Avicel PH 101 in SGF pH 1.2 at 37° C.

Formula	% released after time interval (min)								
	15	30	45	60	90	120	150	180	240
Plain drug	21.48	38.80	47.92	56.12	65.08	72.96	76.48	81.28	88.20
Ph mix 1:1	22.75	53.44	71.31	79.37	87.44	95.08	95.24	96.62	99.88
Ph mix 1:2	12.96	35.24	54.34	70.41	84.94	87.46	93.88	94.75	96.09
Ph mix 1:3	10.51	32.78	48.01	68.15	87.25	93.16	94.36	95.23	97.76
Ad mix 1:1	26.50	35.00	38.86	40.66	43.98	46.85	50.66	51.55	58.02
Ad mix 1:2	41.40	47.77	53.32	60.84	66.60	73.34	76.68	84.72	86.24
Ad mix 1:3	40.39	57.61	70.02	77.75	82.79	86.80	90.70	90.83	95.42

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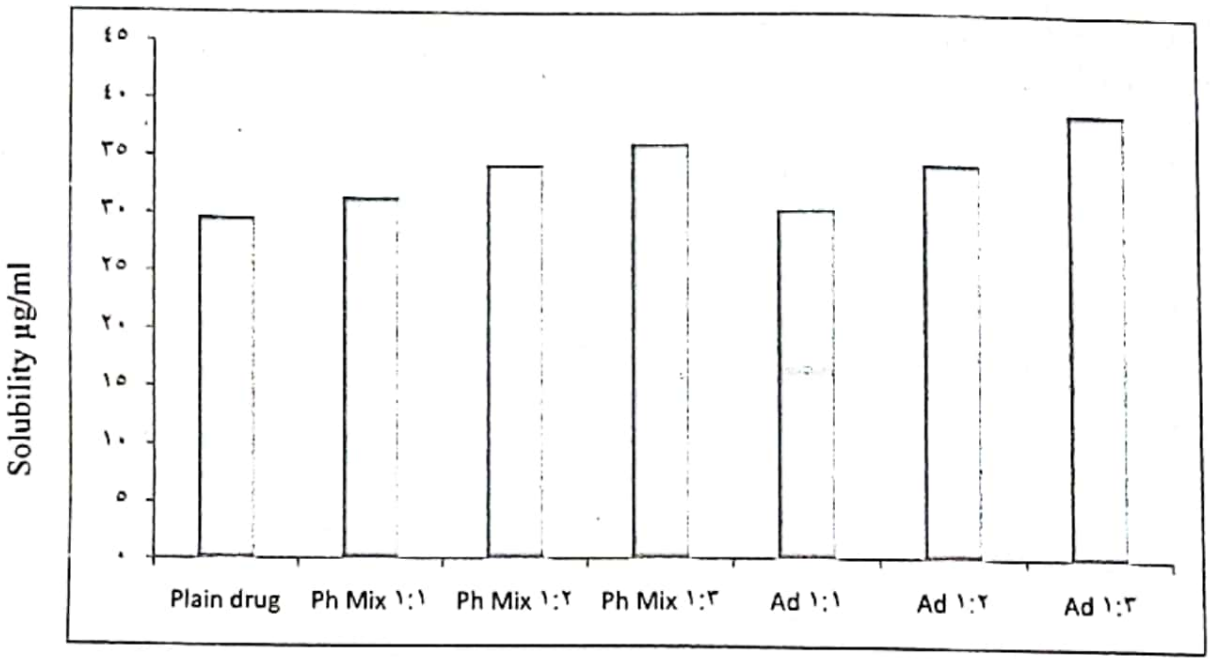

Figure 4. Solubility of spironolactone, its physical mixtures and its adsorbate mixtures in SIF pH 7.5 at 37° C.

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Plain drug	21.48	38.80	47.92	56.12	65.08	72.96	76.48	81.28	88.20
Ph mix 1:1	22.75	53.44	71.31	79.37	87.44	95.08	95.24	96.62	99.88
Ph mix 1:2	12.96	35.24	54.34	70.41	84.94	87.46	93.88	94.75	96.09
Ph mix 1:3	10.51	32.78	48.01	68.15	87.25	93.16	94.36	95.23	97.76
Ad mix 1:1	26.50	35.00	38.86	40.66	43.98	46.85	50.66	51.55	58.02
Ad mix 1:2	41.40	47.77	53.32	60.84	66.60	73.34	76.68	84.72	86.24
Ad mix 1:3	40.39	57.61	70.02	77.75	82.79	86.80	90.70	90.83	95.42

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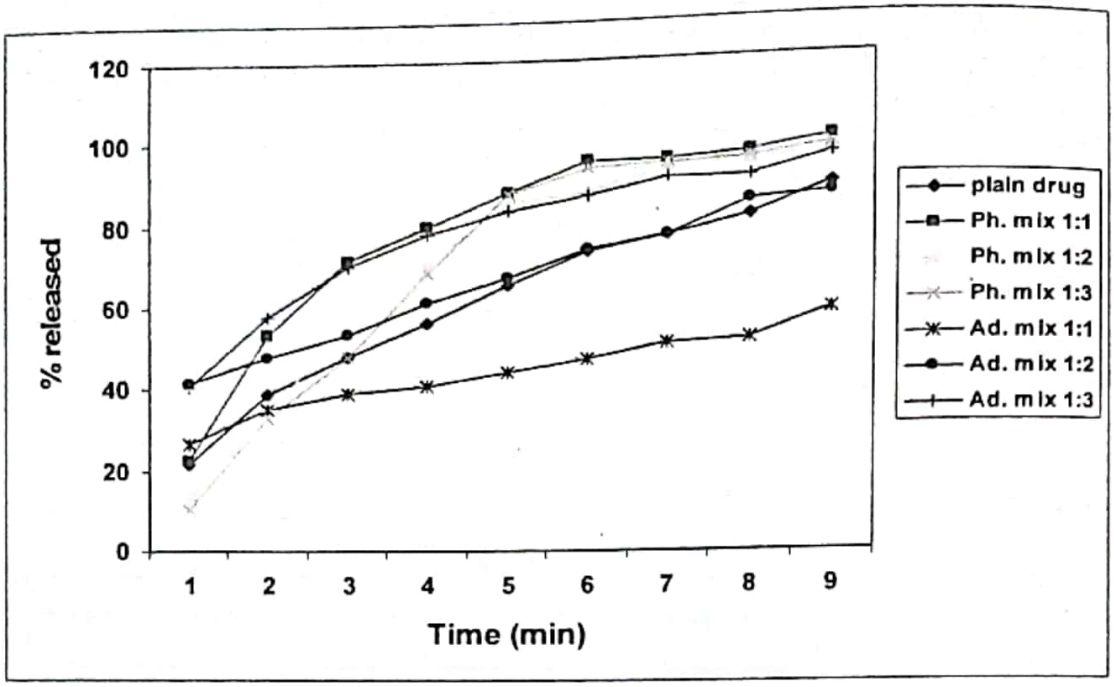

Figure 5. Dissolution profile of spironolactone, its physical mixtures and its adsorbate mixtures with Avicel PH 101 in SGF pH 1.2 at 37° C.

Table (4). *In Vitro* release of spironolactone, its physical mixtures and its adsorbate mixtures with Avicel PH 101 in SIF pH 7.5 at 37° C.

Formula	% released after time interval (min)								
	15	30	45	60	90	120	150	180	240
Plain drug	10.87	35.23	47.88	56.14	69.30	74.69	79.38	84.63	90.92
Ph mix 1:1	6.16	32.83	62.46	72.31	86.60	89.49	93.76	96.18	98.75
Ph mix 1:2	0.29	11.74	33.11	53.39	76.94	86.11	91.05	90.86	96.50
Ph mix 1:3	-	7.60	23.73	44.71	71.99	90.24	92.29	95.89	99.18
Ad mix 1:1	11.99	27.53	34.46	41.16	50.93	55.35	61.73	65.74	72.29
Ad mix 1:2	28.98	43.58	50.87	56.59	65.58	69.64	74.49	75.43	80.63
Ad mix 1:3	0.04	53.06	67.76	72.06	78.65	81.13	85.21	87.04	87.83

- = not determined.

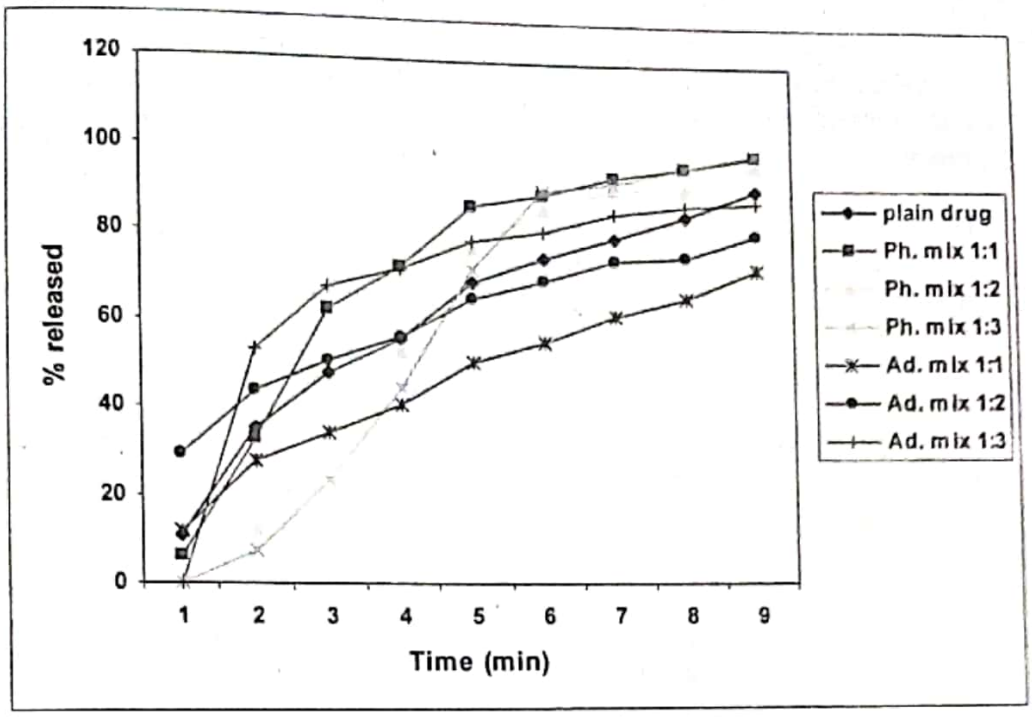

Figure 6. Dissolution profile of spironolactone, its physical mixtures and its adsorbate mixtures with Avicel PH 101 in SIF pH 7.5 at 37° C.

In conclusion, adsorbates of poorly soluble drug spironolactone onto Avicel PH 101 were prepared. The physicochemical charaeterization of the systems prepared showed incomplete transformation of the drug form the crystalline form to the amorphous form with a little improvement in the dissolution pattern of the drug in case of the adsorbates and a more improvement in case of physical mixtures.

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تحضير وتوصيف مخاليط فيزيقية وممتزات من عقار الاسبيرونولاكتون مع الأفيسيل

خالد إسماعيل صالح

قسم الصيدلانيات والصيدلة الصناعية – كلية الصيدلة

جامعة الأزهر – فرع أسيوط

الاسبيرونولاكتون هو عقار ستيرودى يعمل كمضاد خاصة للألدوستيرون ويستخدم كمدر للبول محافظ على البوتاسيوم .

الاسبيرونولاكتون هو عبارة عن مسحوق ذى شكل بلورى ولونه كريمى خفيف وله رائحة شبيهة بالميركابتان ، وهو يعتبر شحيح الذوبان فى الماء ويذوب فى الكحول الإثيلى وخلات الإيثيل ويذوب بدرجة أعلى فى الكلوروفورم والبنزين ويذوب بدرجة قليلة فى الكحول الميثيلى والزيوت الثابتة .

الاسبيرونولاكتون هو عقار شحيح الذوبان فى الماء وله اختلاف فى درجة الذوبان وبسبب قلة ذوبانه ومعدل إتاحته يكون امتصاصه متغيراً أو غير كامل .

قد تم استخدام وتطوير طرق كثيرة لتحسين إتاحة العقارات شحيحة الذوبان فى الماء مثل الاسبيرونولاكتون لتحسين الإتاحة البيولوجية للدواء . من هذه الطرق تصغير الحجم الجزيئى واستخدام المذيبات وتكوين الأملاح وتحضير المشتتات الصلبة من العقار .

وكان الغرض من هذا العمل هو تحضير مخاليط فيزيقية وممتزات من عقار الاسبيرونولاكتون مع الأفيسيل ودراسة تأثير هذا النظام كطريقة لتحسين ذوبان وإتاحة العقار.

وقد أثبتت النتائج أن التحسن فى ذوبان وإتاحة العقار كان أكثر فى حالة المخاليط الفيزيقية عنه فى حالة ممتزات العقار .