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Chemical and Photochemical Stability Studies on Diloxanide Furoate in Carbohydrates and Polyols Solutions

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ABSTRACT

The effects of aqueous solutions containing sucrose, glucose, fructose, lactose and sorbitol on the chemical stability of diloxanide furoate (DF) were investigated in acid and alkaline media at 40˚C and room temperature (25˚C ± 1, adjusted through thermostatically controlled central-cooling system). All above compounds accelerated the rate of hydrolysis of DF in alkaline medium at 40˚C. At room temperature, different brands of sucrose and glucose had adverse effects on the stability of DF. On the other hand, DF was relatively stable in fructose, lactose and sorbitol, as well as in a commercially available Tang® preparation containing sugar, buffers, stabilizers and artificial colours. The optimum pH range of stability of DF in solutions containing these carbohydrates lies below pH 4.5. The influence of β-cyclodextrin (β-CD) and hydroxypropyl β-cyclodextrin (HP-β-CD) on the chemical and photochemical stability of DF was also investigated. *Para*-aminobenzoic acid (PABA), ascorbic acid and the Tang® preparation showed good photo-protective effect on DF solutions irradiated at 254 nm in quartz cell. The observed accelerated rate of hydrolysis of DF by carbohydrates in alkaline medium is proposed to be due to a nucleophilic reaction mechanism while the photostabilization of DF solutions in the presence of PABA, ascorbic acid and the Tang® preparation is proposed to be due to a spectral overlay effect. All above studies were conducted using a stability-indicating HPLC method.

Key words: diloxanide furoate, stability, sugars, cyclodextrins, *para*-aminobenzoic acid, ascorbic acid, HPLC

INTRODUCTION

Diloxanide furoate (DF), 4-(N-methyl-2,2-dichloracetamido)phenyl-2-furoate, is a luminal amoebicide (Figure 1). The drug is formulated in tablet form and orally administered where it is hydrolyzed in the gut to release the active drug, diloxanide[(1)].

We reported the chemical stability (effects of pH, temperature, gastric and intestinal fluids) and photostability of DF using a developed stability-indicating HPLC method in previous studies[(2,3)]. This study investigated the -influence of carbohydrates, polyhydric alcohols, cyclodextrin, *para*-aminobenzoic acid, ascorbic acid and a commercially available Tang® preparation on the stability of DF in aqueous solution. These studies were carried out as a part of the preformulation studies to characterize the physicochemical properties of DF in the presence of these materials. Ideally, the results should help the formulating pharmacist in developing possible formulations of DF in new dosage forms, containing suitable diluents or sweetening agents and packed in suitable containers that can offer optimum stability and longer shelf-life.

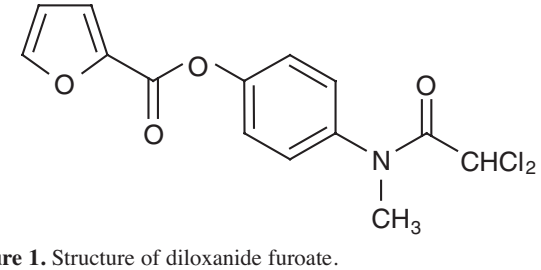

Figure 1. Structure of diloxanide furoate.

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MATERIALS AND METHODS

I. *Apparatus*

A Waters liquid chromatograph consisted of a 600 E system controller, Rheodyne 7161 injector fitted with 20 μL loop, tunable absorbance detector 486 and 746 data module was used. The column employed was 100 RP-18 Lichrosphere (150 × 4.6 mm i.d., 5μm). The mobile phase used was water/acetonitrile (45/55, v/v) filtered through Millipore filter (0.22 μm) and degassed by bubbling helium gas (20 mL min^{-1}) into the solvent reservoir. Afterwards, it was pumped isoractically at a flow rate of 1.2 mL min^{-1}. The UV detector was set at 258 nm, while attenuation was set at 16. Ultraviolet spectrophotometric study was carried out using a Shimadzu UV 1601 PC Spectrophotometer (Kyoto, Japan). The photodegradation process was carried out using a UV-lamp model UVGL-2 (Minerlight® Lamp

multiband UV-254/366 nm, 215-250 volts, 50/60 Hz, 0.12 Amps, San Gabriel, USA) fixed to a wooden cabinet in a horizontal position. pH measurements were done using a Microprocessor pH-Meter BT 500 (Boeco, Germany).

II. *Chemicals and Reagents*

Pure diloxanide furoate (DF) drug sample was provided by Eipico Pharmaceutical Company, Cairo, Egypt. Acetonitrile (Hypersolv^{TM}, BDH, England). Lichrosphere 100 RP-18 (150 × 4.6 mm i.d., 5 μm, Phase Separation Ltd). Buffers: borate, phosphate[4], carbonate[5], and tris buffer[6]. β-CD and HP-β-CD (Fluka Chemika, Switzerland). Ascorbic acid (Fisher Biotech). *Para*-aminobenzoic acid (BDH, England). D(–) fructose and D(+) glucose (BDH, England), sorbitol (Winlab, UK), sucrose (two brands: BDH and Winlab, UK), and two brands of cane sugar (labeled 100% pure) were purchased from the local market. Lactose (Spectrum, New Brunswick), a commercially available Tang® berry flavour (Prod. 27/2/2003; Exp. 27/8/2003; Lot No. 1(1436) (Kraft USA, Kraft Foods, North America, Inc. 800 Westchester Avenue), was obtained from the local market labeled to contain: sugar, citric acid, anticaking agent (Tricalcium phosphate), natural, nature identical and artificial berry flavouring, artificial colours (Allura Red AC, Brillaint Blue FCF), vitamin C, stabilizers (sodium carboxy methyl cellulose, xanthangum) and acidity regulator (trisodium citrate). Quantities were not disclosed on the label. All chemicals were of analytical grade and the reagents used were of laboratory grade.

III. *Procedures*

(I) *Kinetic measurements*

1. Stability Studies in Aqueous Media at Room Temperature (25˚C ± 1)

DF solutions (20 μg mL^{-1} in CH_3CN/H_2O, 55/45 v/v) containing 20% (w/v) of any of the carbohydrate, 2-20% (w/v) of the Tang® preparation or 0.50% (w/v) of β-CD were prepared in aqueous media. The solutions, left in the dark, were analyzed using HPLC method at suitable time intervals. pH values were also measured. A plot of log A_t/A_o *vs* time (t) in days was done where A_t is the area corresponding to the remaining drug at time, t (the duration of study) and A_o is the area corresponding to the drug concentration at initial time.

2. Stability Studies in Aqueous Buffer Solutions at 40˚C

Experiments were carried out at 40˚C for DF solutions (20 μg mL^{-1} in CH_3CN/H_2O, 55/45, v/v) in aqueous buffer solutions (borate, phosphate, carbonate, and tris buffer) each containing varying amounts of the carbohydrates, sorbitol, and 0.50% (w/v) β-CD or HP-β-CD. Volumes of 20 μL were withdrawn at suitable time intervals and analyzed by the described HPLC method. A plot of log A_t/A_o *vs* time was done, where A_t is the area corresponding to the remaining drug at time (t) in hours or minutes and A_o is the area corresponding to the drug concentration at initial time. pH values were also measured.

3. The Influence of Cyclodextrins on the Stability of DF Solution Irradiated at 254 nm in Quartz Cells

DF solution (20 μg mL^{-1} in CH_3CN/H_2O, 55/45, v/v) containing 0.5% (w/v) β-CD or HP-β-CD was placed in a wooden cabinet at a distance of 3 cm from the light source. Irradiation was carried out at 254 nm at different time intervals. The photodegradation was followed up by the developed HPLC method. A plot of log A_t/A_o *vs* time was done and the results were compared with control solution irradiated similarly.

4. Stabilization of DF Solutions Irradiated at 254 nm in Quartz Cell with PABA, Ascorbic Acid and the Tang® Preparation

(1) Stabilization with PABA in aqueous solution (7%, v/v, CH_3CN in Water)

DF solutions (20 μg mL^{-1} in 7% (v/v) CH_3CN in water) containing either 10, 50 or 100 μg mL^{-1} of PABA were prepared. The solutions contained in quartz cells were irradiated at 254 nm and 20 μL volumes were withdrawn at suitable time intervals and analyzed by the HPLC method. A plot of log A_t/A_o *vs* time was done for each solution and results were compared with a control solution irradiated similarly. The half-life values were calculated using the formula, $t_{1/2} = \frac{0.693}{k_{obs}}$ where k is the reaction rate constant (min^{-1}).

(2) Stabilization with PABA in 70% (v/v) CH_3CN in water solution

DF solutions (20 μg mL^{-1} in 70% (v/v) CH_3CN in water) containing either 100, 200, 300 or 400 μg mL^{-1} of PABA were prepared. The solutions contained in quartz cells were irradiated at 254 nm and 20 μL volumes were withdrawn at suitable time intervals and analyzed by the HPLC method. A plot of log A_t/A_o *vs* time was constructed and the results were compared with a control solution irradiated similarly. The half-life values were calculated using the formula, $t_{1/2} = \frac{0.693}{k_{obs}}$ where k is the reaction rate constant (min^{-1}).

(3) Stabilization with ascorbic acid

DF solutions (20 μg mL^{-1} in CH_3CN/H_2O, 55/45, v/v) containing either 40, 100, 200, 400, 600 or 800 μg mL^{-1} ascorbic acid were prepared. The solutions contained in quartz cells were irradiated at 254 nm and 20 μL volumes were withdrawn at suitable time intervals and analyzed by the HPLC method. A plot of log A_t/A_o *vs* time was constructed and results were compared with a control solution irradiated similarly. The half-life values were calculated from the obtained k values.

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(4) Stabilization with a commercially available Tang® powder

DF solution (20 μg mL^{-1} in CH_3CN/H_2O, 55/45, v/v) containing either 2% (w/v) or 20% (w/v) Tang® powder were prepared. The solutions contained in quartz cells were irradiated at 254 nm for 30 min. Percent of the remaining DF was calculated relative to DF content of an aliquot of each solution kept as a control. The results were compared with % DF remaining for DF solution free from Tang® powder irradiated similarly.

RESULTS AND DISCUSSION

It is the concern of the formulating pharmacist to collect enough stability data for a drug intended to be formulated in specific safe, active, and stable pharmaceutical forms with acceptable physical properties (appearance, flavour, palatability, color, etc.) and packed appropriately. On the other hand, it would not be practical for the drug designer to always try to avoid the use of chemically labile or photosensitive groups in new molecules. Thus, it is the job of the pharmaceutical controller to conduct preformulation studies that cover physicochemical properties and stability studies of the drug substance under different conditions, in order to find out the optimal conditions for the drug stability. These data will aid the compounding pharmacist to protect his products from any adverse chemical reactions or photolysis reactions.

Sugars and polyhydric alcohols, such as sucrose, glucose, fructose, lactose and sorbitol, etc, are normally used as sweeteners or diluents for oral liquid formulations to increase palatability, to aid formulation, and to a limited extent, to minimize microbial contamination[6]. Cyclodextrins are cyclic oligosaccharides with a central hydrophobic cavity. They are capable of forming inclusion complexes with a wide variety of drugs by taking up a whole or partial molecule, into their hydrophobic cavity and these complexes can affect the physicochemical properties of the encapsulated drug. These compounds have been used to increase aqueous solubility and stability of drugs[7-10]. Thus we targeted these carbohydrates and cyclodextrins in our investigation to establish their effects on DF, which is available in solid dosage form, in an attempt to present data that can aid the formulation of the drug as a dry powder for reconstitution before use.

I. *Effect of Carbohydrates, the Tang® Preparation and β-CD on DF Stability at Room Temperature*

Figure 2 shows the effect of one of the used brands of sucrose (pH 6.30), glucose (pH 5.23) and β-CD (pH 5.45) on the stability of an aqueous solution of DF containing either of these substances stored in the dark at room temperature for at least 30 days. The results were compared with an aqueous solution of DF (pH 5.50) as control. Sucrose showed the greatest adverse effect on the stability

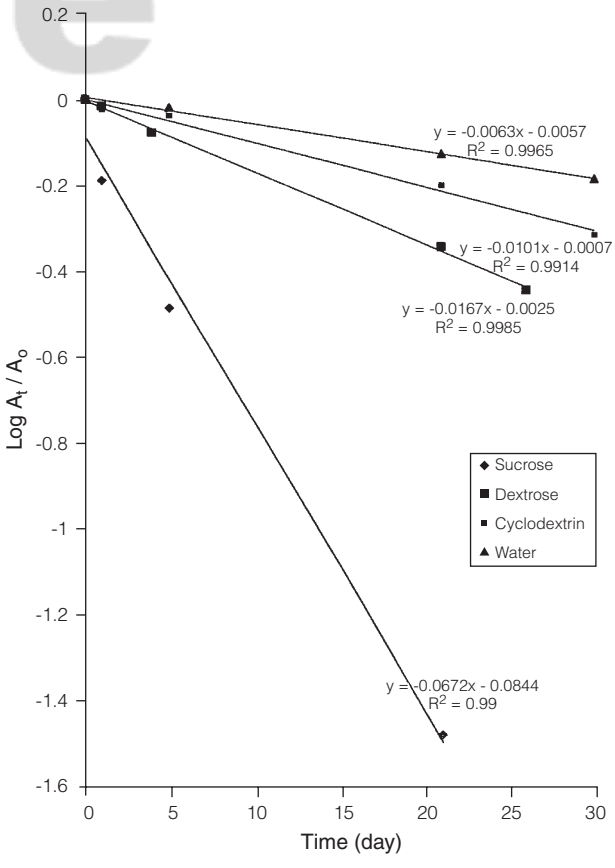

Figure 2. Psuedo-first order plot of the effect of sucrose, glucose, β-CD and water on DF stability. A 30-day study.

of DF leading to rapid hydrolysis within the first few days. The calculated $t_{1/2}$ value for the pseudo-first order plot of log A_t/A_o *vs* time was 4.48 days. The other three brands were found to give varying $t_{1/2}$ values ranging between 3.6 and 9 days. The pH values of these brands ranged between 5.8 ± 0.5. Those with pH values below 5.5 gave more precise $t_{1/2}$ values. Glucose also led to enhanced DF hydrolysis but at a rate slower than that of sucrose; the $t_{1/2}$ value was about 18 days. β-CD showed an even slower hydrolysis rate with $t_{1/2}$ value about 48 days. DF was stable in fructose, lactose and sorbitol (loss in potency was < 8% in one month at pH values < 4.4). In the Tang® preparation, DF was stable for at least 10-month period whether kept in the dark or exposed to daylight (pH ≃ 3.0). The stability of DF in fructose and the comparatively slow rate of hydrolysis by glucose indicate that the effect of sucrose is entirely due to its intact molecule rather than to its hydrolysis products, glucose and fructose. Figure 3 is a typical chromatogram of DF peak (1) in the presence of sucrose in Day 1 (A); (B) from 21 days DF solution in sucrose. The peaks before DF peak are due to its hydrolysis products, diloxanide peak (2) and furoic acid peak (3). The peak before peak (2) was unidentified. However, it was observed for other sugars stored at room temperature for long periods. Chromatogram (c) was from a 21-day old solution of DF in 2% (w/v) Tang® solution showing the intact DF peak (1). The peaks before DF peak are due to

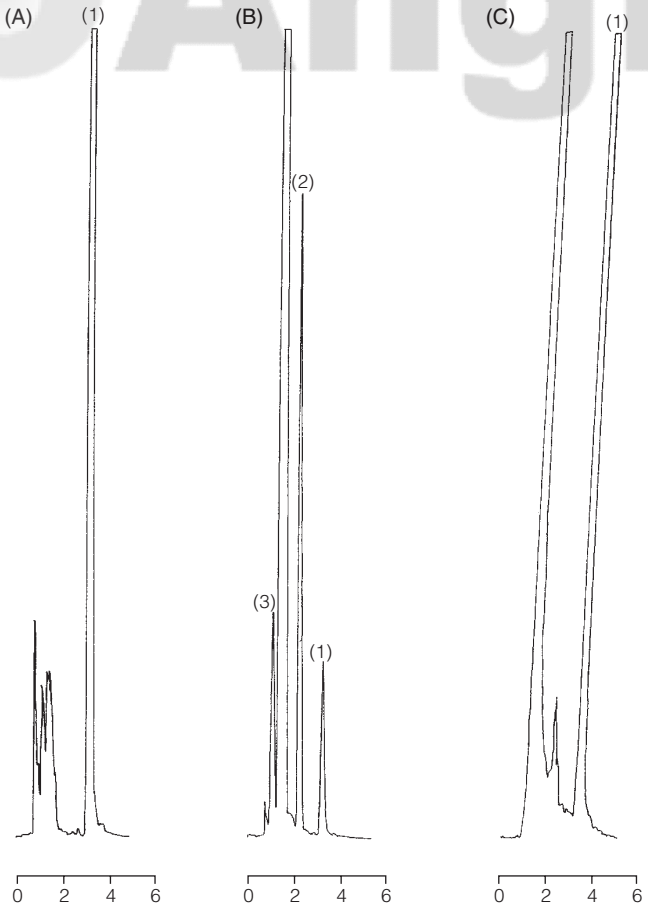

Figure 3. Typical chromatograms (A) DF solution in 20% (w/v) sucrose (day 1). (B) DF solution in 20% (w/v) sucrose (21 days). (c) DF solution in the 2% (w/v) Tang® preparation (day 21). Peak (1), DF intact drug; peak (2), diloxanide; peak (3), furoic acid.

Table 1. $t_{1/2}$ Values obtained from pseudo-first order plots and relative enhancement for the hydrolysis of DF solution (20 μg mL^{-1}) in the absence and presence of carbohydrates and cyclodextrins at different pH and constant temperature 40°C (buffers: borate[(a)], phosphate[(b)], carbonate[(c)] and tris buffer[(d)])

Carbohydrate or β-CD % (w/v)	pH	$t_{1/2}$ (hr^{-1})	Relative enhancement**
None[(a)]	7.4	18.64	1.0
5% Glucose[(a)]*	7.4	3.09	6.0
10% Glucose[(a)]*	6.4	6.60	–
20% Glucose[(a)]*	5.95	11.30	–
5% Lactose[(a)]*	8.30	0.40	
10% Lactose[(a)]*	7.95	0.60	
20% Lactose[(a)]*	7.40	0.79	23.6
None[(a)]	8.60	1.56	1.0
5% Sucrose[(a)]*	8.65	0.24	6.5
10% Sucrose[(a)]*	8.54	0.22	6.5
20% Sucrose[(a)]*	8.26	0.23	–
None[(a)]	9.18	0.67	1.0
0.5% β-CD[(a)]	9.18	0.39	1.72
0.5% HP-β-CD[(a)]	9.18	0.51	1.30
None[(b)]	9.03	1.07	1.0
10% Sorbitol[(b)]	9.03	0.20	5.35
None[(c)]	9.9	0.083	1.0
0.5% β-CD[(c)]	9.9	0.047	1.77
0.5% HP-β-CD[(c)]	9.9	0.058	1.44
None[(d)]	9.06	0.67	1.0
0.5% HP-β-CD[(d)]	9.06	0.63	1.06
0.5% β-CD[(d)]	9.06	0.40	1.70
5% Glucose[(d)]	8.89	0.073	9.2
10% Glucose[(d)]	8.89	0.048	14.0
10% Fructose[(d)]	8.85	0.175	10.0

*Final solutions pH obtained on completing 1 mL of DF solution with borate buffer (0.05 M, pH 9.0) containing the % (w/v) of the nominated sugar.

**Relative enhancement = the ratio between $t_{1/2}$ in the presence and absence of the carbohydrate or β-CD.

some of the Tang® UV absorbing materials observed in the control solution without DF.

II. *Effect of Carbohydrates, Sorbitol, β-CD and HP-β-CD on DF Stability in Aqueous Buffer Solutions at 40°C*

Table 1 shows the $t_{1/2}$ values obtained from the pseudo-first-order plots, and the relative enhancement for the alkaline hydrolysis of DF solution (20 μg mL^{-1}) in the absence and presence of some carbohydrates and cyclodextrins at different pH-values at 40°C. Carbohydrates added to the borate buffer, (0.05 M) pH 9 were observed to cause a large drop in the initial pH value, indicating a low buffer capacity. The pH values of a 10% (w/v) glucose in the buffers used were found to be: 6.4 in borate, 9.55 in carbonate of pH 9.9 (0.2 M) 8.89 in tris buffer pH 9.06 (0.05 M), and 9.0 in phosphate buffer (0.05 M) pH 9.0. All the studied carbohydrates and β-CDs enhanced the hydrolysis rate of DF solution at the pH values shown in Table 1.

As we have reported before[(3)], DF in solutions undergoes decomposition that is temperature and pH dependent. The pH-rate profile at pH in the alkaline side indicated a first-order dependence of k_{obs} on the hydroxyl ions concentration. The pH-rate profile also indicated a high stability of DF in acidic medium. We also suggested a driving effect by transesterification on DF degradation at alkaline pH values in presence of alcohols (methanol, ethanol and *n*-propanol)[(3)]. Thus, the observed driving effect on DF degradation can be attributed to the presence of the hydroxyl groups of these sugars in close proximity to the ester center in DF leading to nucleophilic attack resulting in an accelerated hydrolysis rate. This mechanism was also suggested to be the cause of enhancement of the hydrolysis of β-lactam ring by carbohydrates by an alkoxide ion derived from proton ionization of one of the hydroxyl groups in these compounds[(11)]. Results in Table 1 also shows that the enhancement in the presence of these carbohydrates depends on the [OH^-] and carbohydrate concentration. We also observed that small pH value differences resulted in significant enhancement (at the alkaline pH) or inhibition (at acidic pH). In absence of the carbohydrates, a high catalytic effect of buffer species (at pH values 9-10) is noted for carbonate buffer ($t_{1/2}$ = 0.083 hr) followed by the borate and tris buffer ($t_{1/2}$ for both was 0.67 hr). The phosphate buffer seemed to have the least impact

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on the catalytic process ($t_{1/2}$ = 1.07 hr) but the strongest buffer capacity.

Table 1 results also show the effect of β-CD and HP-β-CD on the stability of DF at pH values of about 9 in borate, phosphate, and tris buffer. β-CD had an adverse effect on the stability of DF showing a relative enhancement in the order of about 1.7-2.0, while the HP-β-CD showed lesser adverse effect. It is known that lower degree of substitution of CDs increases their stabilizing effect as a result of higher complexing ability due to decreased steric hindrance[7]. However, the stabilizing effect of cyclodextrins depends on the nature of the quest molecule, its orientation within the host cavity, and as well as the reaction medium. Thus inclusion complexation can exert an accelerated, decreased or passive effect on the stability of the target drug[7,10]. It is also reported that CDs can accelerate the degradation of β-lactam antibiotics and thus behave like carbohydrates and polyhydric alcohols possessing adjacent hydroxyl groups[7,10]. This last proposal could be the mechanism for the effect of β-CDs on the stability of DF in alkaline medium. Most of the attempts to stabilize drugs with natural CD (β-CD) failed due to their positive catalytic effects on the reactions[9]. Recently, Juleita *et al.*[12] reported stabilizing effect on DF in alkaline medium in the presence of β-CD and its methylated derivatives in the order of 2 and above. In this study we found that β-CD has an accelerating effect on DF hydrolysis rate in alkaline medium in the order of about 2.0. The experimental conditions, such as solvent, temperature, pH and the concentration and the source of β-CD, could be factors that led to this discrepancy in results.

Table 2. Stability of DF solution (20 μg mL^{-1}) in 20% (w/v) sucrose at pH 3.0 and 5.79 stored at room temperature and at 40˚C for 24 hr compared to the Tang® solution pH 3.0. Control solutions were considered 100%

DF in sucrose or Tang®	pH	Temperature (˚C)	% of remaining DF (24 hr)
20% Sucrose	3.0	40˚C	91.48%
20% Sucrose	3.0	25	100.0%
20% Sucrose	5.79	40˚C	51.0
20% Sucrose	5.79	25	81.80
Tang® 5%	3.0	40	100.0%
Tang® 5%	3.0	25	100.0%

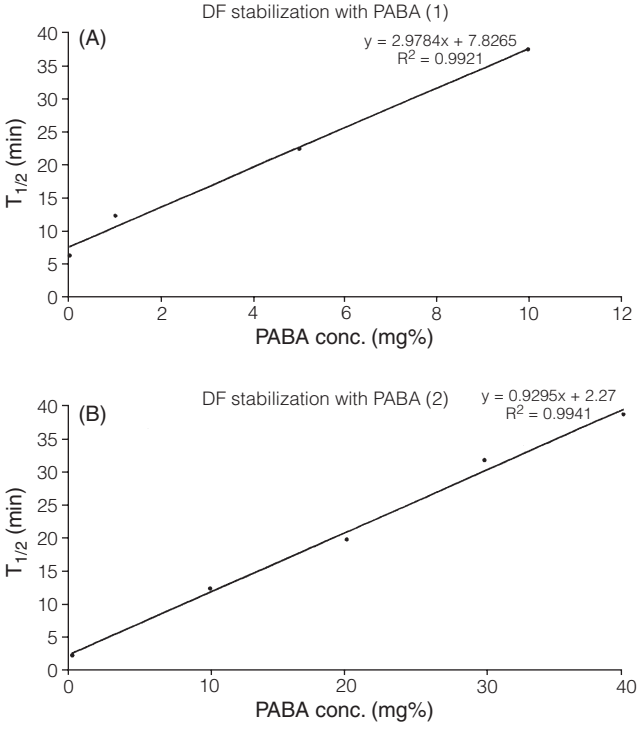

Figure 4. (A) Zero order plot of $t_{1/2}$ values (min) *vs* PABA concentrations (mg%) solution (1). (B) Zero-order plot of $t_{1/2}$ values (min) *vs* PABA concentrations (mg%) solution (2).

The results obtained at room temperature and at 40˚C indicated the high stability of DF in the presence of the various carbohydrates used and in the Tang® preparation at pH values <4.5. To confirm this finding, an experiment was conducted involving the preparation of DF solution in an Analar sucrose either in water (pH 5.79) or in phosphate buffer (pH 3.0) stored at room temperature (25˚C ± 1) and at 40˚C for 24 hr. The results were compared with that of the Tang® preparation (pH 3.0) under the same conditions of temperatures (Table 2). Maximum stability was attained at pH 3.0. In fact, DF in sucrose solution at pH 3.0 remained stable for 8 days without showing any sign of degradation.

III. *Effect of PABA, Ascorbic Acid, Tang® Preparation and Cyclodextrins on the Stability of DF Solutions Irradiated at 254 nm*

Figures 4 (A) and (B) show plots of $t_{1/2}$ values *vs* PABA concentrations for DF solutions in 7% (v/v) CH_3CN in water (4A) and DF solutions in 70% (v/v) CH_3CN in water (4B), respectively. Figure 5 shows a plot of $t_{1/2}$ values *vs* ascorbic acid concentrations for DF solutions in 55% (v/v) CH_3CN in water. Both PABA and ascorbic acid showed good quenching effect on the photochemical reaction of DF with light. Sunscreen-type UV-absorbers (e.g. PABA) have been used as internal protectors for drugs

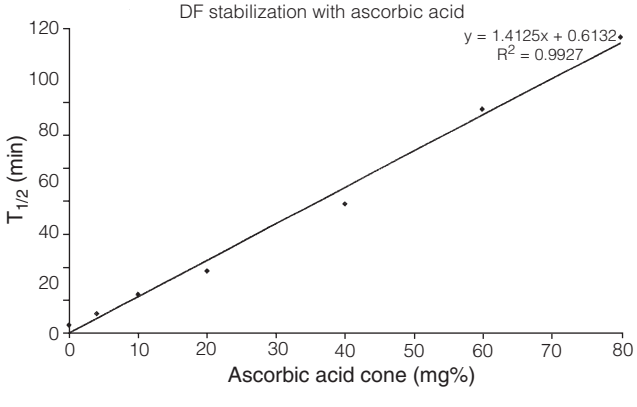

Figure 5. Zero-order plot of $t_{1/2}$ values (min) *vs* ascorbic acid concentrations (mg%).

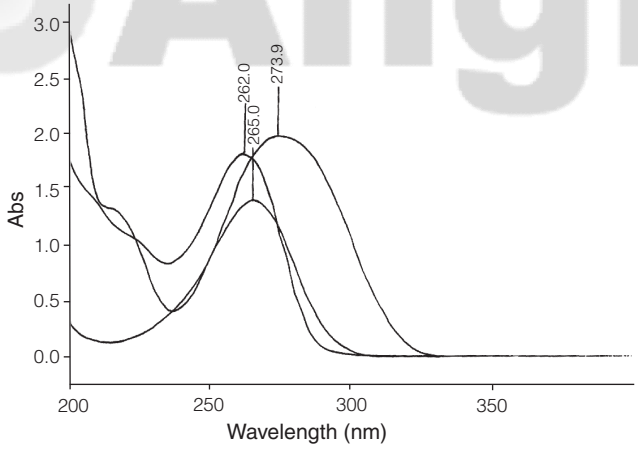

Figure 6. UV-Spectrum of DF (λ_{max} 262 nm); ascorbic acid (λ_{max} 265 nm) and PABA (λ_{max} 274 nm) in aqueous media (solutions 0.002%, w/v).

absorbing at about the same region (spectral overlay or competitive absorption)[13]. Thus it is proposed that the mechanism of the effect of PABA on DF photostabilization could be through a competitive absorption (Figure 6). Lower concentrations of PABA (1-10 mg%, w/v) were needed for good protection for DF solutions in 7% (v/v) CH_3CN in water, whereas 10-40 mg% (w/v) PABA concentrations were needed to protect DF solutions in 70% (v/v) CH_3CN in water. No significant shift in λ_{max} of PABA in both media was observed (λ_{max} = 274 nm). As we reported before[2], the stability of DF to light can be improved by adding water to the organic solvents used to keep the drug in solution. We also observed that DF stability to light is decreased in organic solvents due to possible formation of free radicals that may participate in the photodecomposition reaction. Thus this finding seems to support our previous suggestion about the effect of high % content of organic solvents in the photostability of DF in solution.

The effect of the reducing agent, ascorbic acid, which is used as an antioxidant was thought to be through the quenching of a free radical reaction of light with DF. However, solutions bubbled with nitrogen gave about the same results relative to control solutions within 6% variation. The average DF percentage remaining for solutions irradiated for 5.0 min at 254 nm was 38.45% ± 1.72 (n = 2) for nitrogen saturated solutions, compared to 31.54% ± 0.96 (n = 2) for control solutions and 94.8% ± 1.05 (n = 2) for solutions containing 25 mg% ascorbic acid. These results may not entirely eliminate the possibility of the involvement of minor oxidative reaction, but relative to the non-oxidative effect, this may not be significant. The λ_{max} of an aqueous solution of ascorbic acid is at 265 nm and that of DF is at 262 nm. Thus a spectral overlay effect could be the mechanism of the protecting effect of ascorbic acid (Figure 6). DF solutions containing 0.5% (w/v) β-CD or HP-β-CD irradiated at 254 nm in quartz cells gave about the same $t_{1/2}$ values (6.5 ± 0.4 min) as compared to a control solution indicating no protective effect. On the other hand, DF solutions containing either 2% (w/v) or 20% (w/v) Tang® preparation irradiated at 254 nm in quartz cells for 30 min showed good DF stability. The remaining DF % for the control solution was 7.5% (w/v) compared to 34.5% in 2% (w/v) Tang® and 88.8% for DF solution in 20% (w/v). This high stabilizing effect by the Tang® preparation under stress condition could be attributed to the presence of ascorbic acid and the natural colouring materials which are known as drug stabilizers against photochemical reactions through spectral overlay effect[13].

CONCLUSIONS

DF solutions containing carbohydrates (sucrose, glucose, fructose, lactose) or polyhydric alcohol, sorbitol, have the maximum stability at pH 3.0. *Para*-aminobenzoic acid, ascorbic acid and a commercial Tang® preparation can be good photostabilizers for DF in solution. A transesterification mechanism has been suggested for the effect of the carbohydrates on DF solutions at alkaline pH's and an overlay effect has been proposed to explain the protecting effect on DF solution observed for *para*-aminobenzoic acid and ascorbic acid. However, both mechanisms require further verification. The results obtained in this study coupled with our previous studies on DF stability can be a good guide for the manufacturer or pharmacist if an oral preparation of DF is to be prepared. We recommend the use of the Tang® preparation as effective diluent which can act as flavouring, colouring, stabilizing and sweetening agent.

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