ORIGINAL ARTICLE

CEPHRADINE ANTACIDS INTERACTION STUDIES

M. SAEED ARAYNE, NAJMA SULTANA*, AND M. AFZAL**

Department of Chemistry, University of Karachi, Karachi-75270, Pakistan

**Department of Pharmaceutical Chemistry, Research Institute of Pharmaceutical Sciences, Faculty of Pharmacy, University of Karachi*

***Military College, Jehlum, Pakistan*

ABSTRACT

The present work comprises of interaction studies of cephradine with antacids. Cephradine is included among the first generation cephalosporin, which is active against a wide range of Gram positive and Gram-negative bacteria including penicillinase-producing staphylococci. Since the presence of complexing ligand may affect the bioavailability of a drug in blood or tissues, therefore, in order to study the probable interaction of cephradine with antacids all the reaction conditions were simulated to natural environments. Antacids are commonly used in patients complaining of GI irritations. The behavior of cephradine in presence of seven antacids i.e., simethicone, magaldrate, magnesium carbonate, magnesium hydroxide, magnesium trisilicate, sodium bicarbonate and aluminium hydroxide was studied by using standard dissolution apparatus. Cephradine was monitored both by UV and by high performance liquid chromatography. The results revealed that antacids containing polyvalent cations retarded the *in vitro* availability of cephradine. Moreover, these studies indicated that cephradine was strongly adsorbed on antacids; magnesium trisilicate and simeco® tablets (powdered) exhibited relatively higher adsorption capacities.

Keywords: Cephradine; antacids; simethicone; magaldrate; magnesium carbonate; magnesium hydroxide; magnesium trisilicate; sodium bicarbonate; aluminium hydroxide.

INTRODUCTION

Cephradine, the semisynthetic first generation cephalosporin has shown good activity against Gram-positive bacteria. Like other members of family it has clinical significance. It is a white or slightly yellow, hygroscopic powder, sparingly soluble in water, practically insoluble in alcohol, diethyl ether, chloroform, benzene and hexane, very slightly soluble in acetone (BP 2005). Cephradine melts with decomposition at 183-185°C whereas it has a varied range from 175-192°C.

Corresponding author: E-mail: arayne@gawab.com

Various methods are reported in the literature for the assay of cephradine [Campos *et al*., 1993; Craft and Forster 1978; Hayashi 1981; Kai *et al*., 2003; Kunst and Mattie 1978; Nunez-Vergara *et al*., 1979; Nunez-Vergara *et al*., 1991; Ryu *et al*., 1977; Warren *et al*., 1978; Welling *et al*., 1979; Zhu *et al*., 1994]. On the other hand there are number of methods reported for the simultaneous analysis of cephradine from other cephalosporins (Wu *et al.,* 1999), human plasma (Johnson *et al*., 2000; McAteer *et al*., 1987) and urine (Clarke and Robinson, 1983) by HPLC with UV detection. Moreover, Cephradine has also been estimated from aqueous solutions (Wang and Monkhouse, 1983) and solutions

containing sodium bicarbonate (Yamana and Tsuji, 1976). In our studies cephradine was estimated from reference drug as well as antacid containing solutions both by HPLC with UV detection as well as directly by UV detection at 254 nm.

Antacids are drugs taken orally to neutralize stomach acidity. They temporarily raise the pH of gastric contents and relieve the pain of peptic ulcer. Two types of antacids were used in our studies systemic (highly soluble and are rapidly absorbed from the gut and may cause systemic alkalosis) which includes sodium bicarbonate and nonsystemic (less soluble and exert their action locally in G.I. tract producing no drastic changes in acid-base balance) which includes magnesium trisilicate, magnesium oxide and magnesium hydroxide, aluminum hydroxide and magaldrate, calcium carbonate and hydroxide.

Keeping in view the fact that bioavailability of cephalosporins at their site of action can be enhanced or reduced by interaction with other drugs (Cathcart *et al.,* 1989; Masroor 1997), present work was planned to study the availability of cephradine in presence of antacids and to quantitate cephradine-antacid interactions.

MATERIALS AND METHOD

Materials

Velosef and Valodin (Cephradine) were gifts from Squibb Laboratories Karachi and Hilton Pharma Ltd Karachi. These materials had expiry date not earlier than 365 days, at the time of these studies. All the reagents used were of analytical grade or purified in the laboratory according to standard procedures (Brain *et al.,* 1989). Magaldrate, aluminum hydroxide, sodium bicarbonate, magnesium hydroxide, calcium carbonate were of pharmaceutical grade. Deionized water was used through out the work. All the glasswares were washed with chromic acid followed by a thorough washing with water and finally rinsed with double distilled or deionized water, which was freshly prepared in the laboratory.

Methods

The cephradine antacid interaction studies were carried out on dissolution test equipment as it simulates the alveolar movements of the stomach. The dissolution test equipment was manufactured to the Pharmacopoeial standards (B.P. 2005), with slight medications (Sarapu and Clark 1980; Fifth Supplement to USPXIX and NF XIV 1979; Embil and Torosian 1979), details of which have already been reported (Iftikhar *et al.,* 2005). The dissolution assembly was immersed in a water bath at 37 $\pm 0.1^{o}$C. The absorbance of cephradine in the dissolution medium was measured by UV/Visible spectrophotometer or alternatively, the drug was quantitated by HPLC.

Monitoring of cephradine by UV-Visible spectrophotometry

The UV/Visible spectrophotometer comprised of Shimadzu Japan Model 1601, which was serially connected to a P4 computer loaded with UVPC ver 3.9 software. The UV spectrophotometric method selected was simple, accurate and quick. Cephradine exhibited strong absorption in the ultraviolet region of the spectrum at 254 or 262 nm depending upon the pH of the medium. Measurement of this absorption band has been employed for the assay of cephradine, which followed Beer's Lambert Law in the concentration range of 0.01-0.2 mMoles. All the antacids used in these studies did not absorb in the UV or Visible region.

Monitoring of cephradine by RP-HPLC

The cephradine was also simultaneously analyzed on a reverse phase high performance liquid chromatography. The chromatographic system was a Shimadzu HPLC system (Japan), consisted of a LC-10AT VP pump, a SIL-AD VP auto injector and a SPD-10AT VP variable wavelength UV detector. The chromatographic data was integrated using a CBM-102 communication bus module Shimadzu on an IBM PC loaded with CLASS-GC software. Chromatographic separation was carried out at room temperature with a Technoman ODS C18 (200 mm x 4.6 mm i.d., 5 microns) column. The mobile phase MeOH :water (30 :70) was pumped at a flow rate of 1 per ml minute and the drug was monitored at 265 nm.

In vitro availability of cephradine

In the first step, *in vitro* availability of cephradine reference standard was studied without antacids in simulated gastric juice at 37^{o}C on a modified B.P. 2005 dissolution apparatus described earlier. 50 mg of drug was poured in 1-liter dissolution medium (0.1 N HCl) already maintained at specified temperature at the start of the experiment. Aliquots were withdrawn periodically at fifteen minutes time intervals for 180 minutes and assayed for the drug contents. The volume of dissolution fluid was maintained by adding an equivalent amount of dissolution fluid withdrawn, which had previously been maintained at the same temperature in the same bath. The sample was scanned in the region 250-350 nm against reagent blank and the maxima was observed at 254 nm.

Interaction with antacids

In vitro interaction studies of cephradine with each of the antacids was carried out in 0.1 M HCl in the same manner. In these sets of experiments 100 mg of cephradine was added to the dissolution medium at zero time while after 15 minutes before collecting the sample, 2 gm of each antacid was added to the dissolution medium separately in each individual experiment. Aliquots were withdrawn every 15 minutes till three hours and assayed as before. Graphs between cephradine

Table 1: Concentration of cephradine (%) in presence of antacids at different time intervals

Sample	Time (min)											
	5	10	15	20	25	30	35	40	45	50	55	60
Cephradine	15.5	29.8	45.1	65.0	82.0	93.7	100.0	100.6	100.0	99.5	100.6	100.0
Cephradine + Aluminium hydroxide	-	09.3	15.6	21.8	25.0	28.1	31.2	34.3	37.5	40.6	43.7	46.0
Cephradine + Aluminium trisilicate	-	16.4	28.0	35.6	42.8	57.6	63.8	69.5	69.5	70.0	69.6	70.0
Cephradine + magnesium oxide	-	15.6	37.5	60.0	68.7	75.0	81.2	81.2	81.0	80.5	80.0	80.0
Cephradine + magnesium trisilicate	5.2	13.6	9.5	11.4	12.5	13.5	13.8	14.16	14.0	13.9	11.5	14.2
Cephradine + sodium bicarbonate	-	28.1	45.0	63.1	71.8	78.0	81.9	87.5	90.0	92.6	94.8	96.8
Cephradine + SimecoR	6.0	6.2	7.6	8.2	8.6	9.0	10.6	11.1	12.5	12.5	12.5	12.5

concentration versus time were plotted which showed drug status during and at the end of the experiments.

Adsorption studies

The desired quantities of antacid powders was weighed accurately in 25 ml Erlenmeyer flasks. Aqueous solution of cephradine 10 ml at appropriate concentration was added to each flask. The flasks were shaken in a constant temperature bath at 37°C for one hour. It had been established previously that equilibrium was attained within this period. At the end of this time aliquots were filtered through a millipore filter (0.2μ) and analyzed for the residual antibiotic content. The quantities of cephradine adsorbed were calculated by subtracting the equilibrium concentration from the initial concentration. No difference in concentration was found in sample to which antacid had not been added.

RESULT AND DISCUSSION

When cephradine was determined, there was no difference between the results obtained by using HPLC-UV and direct UV measurements. The direct UV measurements give instant results and in HPLC-UV method samples were resolved within fifteen minutes; while the later method was more sensitive and accurate. The correlation coefficient was 0.9999, while LOQ was 107 ng - 10.702 μg and percent recovery was 99.7. The precision of the method at 0.2 μg/ml was 4.9% (intra-assay) and negligible (inter-assay) as calculated by one-way analysis of variance and the accuracy of the method at 0.2 μg/ml was -4.1% in terms of percentage bias.

The results of the effect of antacids on the *in vitro* availability of cephradine at different time intervals are mentioned in table 1. A graph was plotted (fig. 1) for the first order dissolution rate constant of drug concentration versus time in each set of experiment in presence or absence of antacids. The dissolution time T_{50} and T_{90} of cephradine in presence of various antacids are given in table 2. As can be seen from these profiles, the dissolution rate of cephradine decreased in the presence of all the antacids studied except sodium bicarbonate in which case it increased significantly.

In case of sodium bicarbonate there was an increase in the dissolution in first 20 minutes and then there was a decline so that almost an equivalent amount of drug was dissolved as compared to control at the end of the experiment. In our studies when sodium bicarbonate was added to the dissolution medium (0.1N HCl), the concentration of hydrogen ion decreased due to the evolution of CO_2, which led to increase in pH. The pH study of 0.2% solution of sodium bicarbonate in distilled water and 0.1N hydrochloric acid compared with the pH of distilled water and 0.1N hydrochloric acid (table 3) showed that pH may be responsible for increased rate of dissolution as compared with other antacids (Mann, 1972).

Magnesium oxide, which is soluble in 0.1N hydrochloric acid, exhibited an insignificant effect on the rate of dissolution of cephradine. After an interval of 30 minutes 81% of the drug was present in the solution, which was consistent up to 60 minutes. The T_{50} and T_{90} values of cephradine in presence of magnesium oxide were found to 24.76 and 32.38 minutes respectively.

Aluminum trisilicate, which is insoluble in the dissolution medium, exhibited a significant retardation effect on the dissolution of cephradine. After an interval of 30 minutes only 57% and after 60 minutes 70% of the drug was

dissolved so that T50 and T90 values for cephradine in presence of aluminum trisilicate were 26.02 and 84.92 minutes respectively.

From the results as listed in table 1 and 2, it is quite clear that the dissolution rates of cephradine decreased in presence of aluminum hydroxide and magnesium trisilicate in a concentration of 0.2 % W/V. Miyazaki *et al*., (1981) studied the effect of antacids on the dissolution of minocycline and demethylchlortetracycline from capsules. We earlier (Arayne and Sultana 1993) showed that dissolution of erythromycin was found to be markedly retarded in presence of various antacids including aluminium hydroxide and magnesium trisilicate. Similar results were obtained in our studies and presence of even lower levels of antacid (0.2% W/V magnesium trisilicate), also reduced cephradine dissolution; the amount dissolved after 30 minutes were 13.2 and after 60 minutes 14.24 % with a very high T_{50} and T_{90} values.

In case of aluminum hydroxide only 28.12% of the drug was present in the dissolution medium after an interval of 30 minutes and 46% of the drug was present at the end of the experiment with T_{50} and T_{90} values of 64.48 and 135.58 minutes respectively.

When aluminum hydroxide and magnesium trisilicate were added to the dissolution medium, they remained in suspended and undissolved state. There are two possibilities for the slow rate of dissolution, either due to increase in pH or adsorption properties of these two antacids. From the pH studies it is quite clear that pH was not a major factor for prolonged dissolution behavior. Chelating effect of cephradine with Mg^{2+} and Al^{3+} may be responsible for the prolonged and incomplete dissolution.

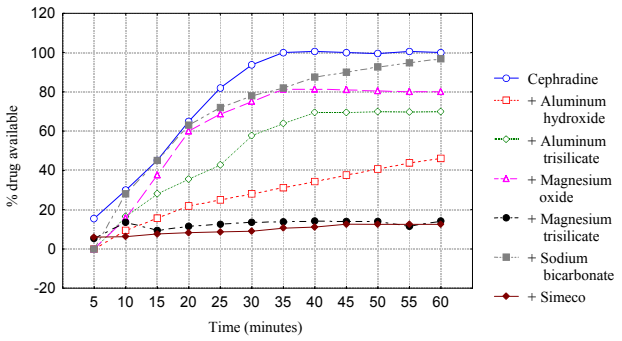

Fig. 1: *In vitro* availability of cephradine in presence of antacids.

Simeco® tablet, a combination of aluminum hydroxide, magnesium carbonate, magnesium hydroxide and methylpolysiloxane had the greatest retardation effect on

Table 2: Dissolution rate constants of cephradine in presence of antacids

Sample	$T_{50\%}$	$T_{90\%}$	K
Cephradine	23.09	28.04	0.4199
Cephradine + Aluminium hydroxide	64.48	135.58	2.92 x 10-2
Cephradine + Aluminium trisilicate	26.02	84.92	5.2 x 10-2
Cephradine + magnesium oxide	24.76	32.38	0.2726
Cephradine + magnesium trisilicate	125.70	294.47	1.23 x 10-2
Cephradine + sodium bicarbonate	32.06	51.23	0.1078
Cephradine + SimecoR	202.22	493.43	7.13 x 10-3

Table 3: Adsorption capacities towards cephradine and pH behavior of antacids in water and in 0.1N HCl

Antacid	pH in*		Adsorption capacity** [mg(mmoles)/gm] for cephradine
	H_2O	HCl	
Aluminium hydroxide	7.60	1.25	270 (0.367)
Aluminium trisilicate	9.7	1.6	153 (0.208)
Magnesium oxide	10.70	1.85	100 (0.136)
Magnesium trisilicate	9.60	1.37	429 (0.584)
Sodium bicarbonate	8.73	1.33	15 (0.02)
SimecoR #	1.5	1.2	438 (0.596)

pH of distilled water = 7.00; pH of 0.1N HCl = 1.10; * = antacid concentration 0.2% W/V;
** = antacid concentration 1% W/V; # = 2 tablets (850 mg) for each study
R = Registered trade mark of Sterling Winthrop Ltd.

the dissolution behavior of cephradine. Only 9.04% and 13% of the drug was dissolved after 30 and 60 minutes respectively so that T_{50} and T_{90} for cephradine in presence of Simeco® tablets were 202.22 and 493.43 minutes.

Thus it is clear that the dissolution of cephradine can be retarded by small amounts of antacids containing polyvalent cations. Although it had previously been suggested that antacids decrease the dissolution of other antibiotics by raising the pH of the medium (Sultana *et al*., 1983; Sultana *et al.,* 1984) and the dissolution rate is markedly reduced at high pH values (Gugler and Allgayer 1990), there was no significant increase in pH by the addition of these antacids in the dissolution medium. Table 3 shows the pH of the antacids in distilled water and in 0.1N HCl. On the other hand cephradine was found to be strongly adsorbed on various antacids. Figure 2 shows the data plotted according to the following equation.

$$\frac{c}{x/m} = \frac{1}{ab} + \frac{c}{b}$$

Where "c" is equilibrium concentration of the solute, x/m is the amount of solute adsorbed per unit weight of the adsorbent, and "a" and "b" are constants. The adsorption capacities of the antacids are also listed in table 3. Magnesium trisilicate and Simeco® exhibited relatively higher adsorption capacities.

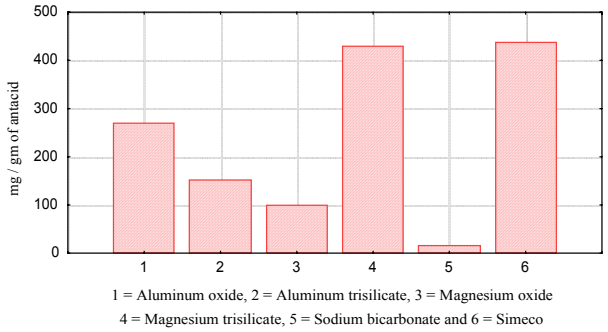

Fig. 2: Adsorption capacities of various antacids towards cephradine.

CONCLUSION

It is thus clear that antacids containing polyvalent cations can retard the dissolution of cephradine. These studies indicated that cephradine is strongly adsorbed on antacids; magnesium trisilicate and simeco® tablets (powdered) exhibited relatively higher adsorption capacities. The difference in the adsorption capacities could not be related to the pH of the antacids suspensions. The adsorption of cephradine by antacids may be responsible for the marked retardation of dissolution and hence the absorption of cephradine. Similar mechanisms were reported for the inhibited dissolution of glycosides (Domenico *et al*., 1991; Hooymans and Merkus, 1986; Sonobe *et al*., 1980), contraceptive steroids (Khalil and Iwuagwu, 1976) and tetracyclines (Sultana *et al*., 1983; Sultana *et al*., 1984). Chelation may also be considered to be one of the mechanisms responsible for the decreased absorption of antibiotics in presence of antacids containing di and trivalent metal cations (Spratt and Cromie, 1988). There have been number of reports on the repressed bioavailability of other antibiotics in presence of minerals and trace elements administered or present in the body (FrŠre and Joris, 1985; Ghuysen 1977; Ghuysen, 1984).

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