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The development of a stable oral solution of captopril for paediatric patients

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Abstract

Study objectives: Many major drugs are not available in paediatric form. The aim of this study was to develop a stable liquid solution of captopril for oral paediatric use allowing individualised dosage and easy administration to newborn and young patients.

Methods: A specific HPLC-UV method was developed. In a pilot study, a number of formulations described in the literature as affording one-month stability were examined. In the proper long-term study, the formulation that gave the best results was then prepared in large batches and its stability monitored for two years at 5°C and room temperature, and for one year at 40°C.

Results: Most formulations described in the literature were found wanting in our pilot study. A simple solution of the drug (1 mg/mL) in purified water (European Pharmacopeia) containing 0.1% disodium edetate (EDTA-Na) as preservative proved chemically and microbiologically stable at 5°C and room temperature for two years.

Conclusion: The proposed in-house formulation fulfils stringent criteria of purity and stability and is fully acceptable for administration to newborn and young patients.

Keywords

Captopril, formulation, pharmaceutical technology, chemical stability, microbial stability, high-performance liquid chromatography (HPLC)

Introduction

According to recent studies, a significant proportion of medicines prescribed to hospitalised newborns and children do not exist as paediatric formulations [1, 2]. This does not imply that the prescribed drug is ineffective or unsafe in such patients, but more simply, that manufacturers have no incentive to develop such a form.

This situation presents hospital pharmacists with a clear challenge. Given the impossibility of administering tablets or capsules to newborns or infants, a liquid formulation has to be developed whose stability must be optimised and shelf-life determined. This paper describes such a study involving the oral formulation of captopril.

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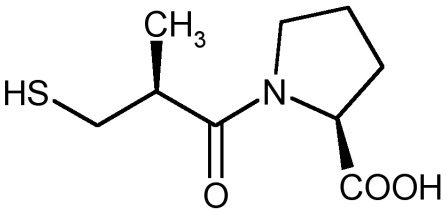
Figure 1: The structure of captopril



Captopril (Figure 1) is a well-known inhibitor of angiotensin-converting enzyme (ACE) frequently used to treat arterial hypertension and congestive cardiac failure in adults and children [3-5]. A comprehensive study carried out in 1997 in 53 French hospitals has shown that the most frequently prescribed paediatric drugs were (in decreasing order): diphemanil, captopril, fludrocortisone, ranitidine, spironolactone and ursodeoxycholic acid [6]. Initial doses of captopril administered to premature neonates and newborns range from 0.01 to 0.1 mg/kg every eight to 24 hours. The dose has then to be titrated up to a maximum of 0.5 mg/kg/dose given every six to 24 hours [3].

Whereas the hydrolytic cleavage of captopril is negligible

under pharmaceutically relevant conditions, its oxidative dimerisation to a disulphide is a significant pharmaceutical problem. The reaction is catalysed by metal ions and its rate depends on pH and oxygen concentrations. Thus, the oxidation of captopril is lowest at pH 4, and is markedly slowed down by chelating agents, antioxidants, high concentrations of the drug, and a small and nitrogen-saturated headspace [7-11].

A number of studies have investigated the stability of captopril in boiled tap water, distilled water, sterile water, diluted syrup containing 2% methylcellulose, and in the oral vehicles Ora-Sweet, Ora-Sweet SF and Ora-Plus. EDTA-Na and ascorbic acid were also used as additives. The stability of captopril in these preparations was highly variable and depended on the quality of the raw materials used [12-24]. In a one-month study, the most stable sample of captopril was obtained with a pH 3.2 solution containing EDTA-Na and stored at 5°C in flasks of brown glass [20].

The objective of our study was to obtain a captopril solution suitable for oral administration to newborns and young children and showing very good chemical and microbiological stability. We have already reported some preliminary results [25].

Materials and methods

Chemicals

Water complying with European Pharmacopeia (Pharm Eur) standards, obtained by reverse osmosis, was used [26]. Captopril Pharm Eur (produced by BUFA BV Pharmaceutical Products) was supplied by Dynapharm (Meyrin, Switzerland). Ascorbic acid Pharm Eur was purchased from Hänseler (Herisau, Switzerland) and EDTA-Na Pharm Eur from Merck (Dietikon, Switzerland). Oral-Plus and Ora-Sweet (produced by Paddock Laboratories) was obtained from Nolte (Innenburen, Germany). Oral-Plus is an oral suspending vehicle containing viscosifying agents (cellulose, carboxymethylcellulose, xanthan gum and carrageen), buffers (citric acid and sodium phosphate), an antifoam agent (simeticone), and preservatives (methylparaben and potassium sorbate). Ora-Sweet is a syrup vehicle containing sucrose, glycerin and sorbitol, buffers (citric acid and sodium phosphate), and preservatives (methylparaben and potassium sorbate).

All analytical solvents and reagents were of HPLC grade. Phosphoric acid 85%, water and acetonitrile were obtained from VWR International (Dietikon, Switzerland). Brown flasks (Veral) were obtained from Mueller and Krempel SA (St-Prex, Switzerland).

Analytical methods

The concentration of captopril was measured by HPLC as described in the literature [12, 15, 16, 19]. A Varian automated HPLC system with StarStation software was used consisting of a 9012 pump, a Prostar 410 autosampler, an incorporated column oven (temperature range from room temperature to 60°C, accuracy: ± 1°C), a 9065 diode-array (DAD) detector and a computer. The stationary phase was a Hamilton PRP-1 analytical column (150 x 4.1 mm, 5 µm particle size, 100 Å pore size) heated to 50°C. The mobile phase was a 77:23 v/v mixture of phosphoric acid 0.01 M and acetonitrile. Samples were diluted 1:10 with phosphoric acid 0.01 M, and the injection volume was 20 µL. Flow rate was 1.0 mL/min, and measurements were made at 205 nm. The retention time of captopril was three minutes.

The forced degradation of captopril in solution (1 mg/mL) showed the HPLC assay to be stability-indicating and allowed its validation according to the guide published by the Société Française des Sciences et Techniques Pharmaceutiques [27]. The five distinct conditions for forced degradation were as follows:

1. Heat degradation of the unbuffered solution of captopril at 100 ± 1°C for one hour.
2. Acidification of the captopril solution with the same volume of a hydrochloric acid (HCl) 5N solution, then storing for one hour at room temperature. This acidic solution was neutralised with sodium hydroxide (NaOH) 2N before analysis.
3. Alkalinisation of the captopril solution with the same volume of a NaOH 5N solution, then storing for one hour at room temperature. This alkaline solution was neutralised with HCl 2N before analysis.
4. Mixing of the captopril solution with the same volume of a sodium peroxide (H_2O_2) 0.3% solution, then storing for one hour at room temperature.
5. Exposure of the captopril solution to daylight for a total of 30 hours.

In the chromatograms obtained from these tests, captopril was always well separated from its degradation products (see later). External standard curves were produced for each assay using five dilutions of the solution (0.06-0.14 mg/mL).

Other tests

Microbiological tests were carried out at the beginning, after

six months and at the end of the long-term study according to the protocol of the European Pharmacopeia 4th Edition [28]. The pH was checked at regular intervals using a Metrohm 713 pH-meter (Metrohm, Herisau, Switzerland). Organoleptic tests included clarity (observation against a black background) and opalescence (observation against a white background), with purified water as control. An olfactory test was also performed.

Pilot study

A pilot study of one month's duration comprised a first phase. Ten aqueous formulations of captopril 1 mg/mL (Table 1) were prepared, and their physical and chemical stability compared after one month. Similar formulations are described in the literature as having a maximal stability of one month at room temperature [16-21, 24].

Long-term study

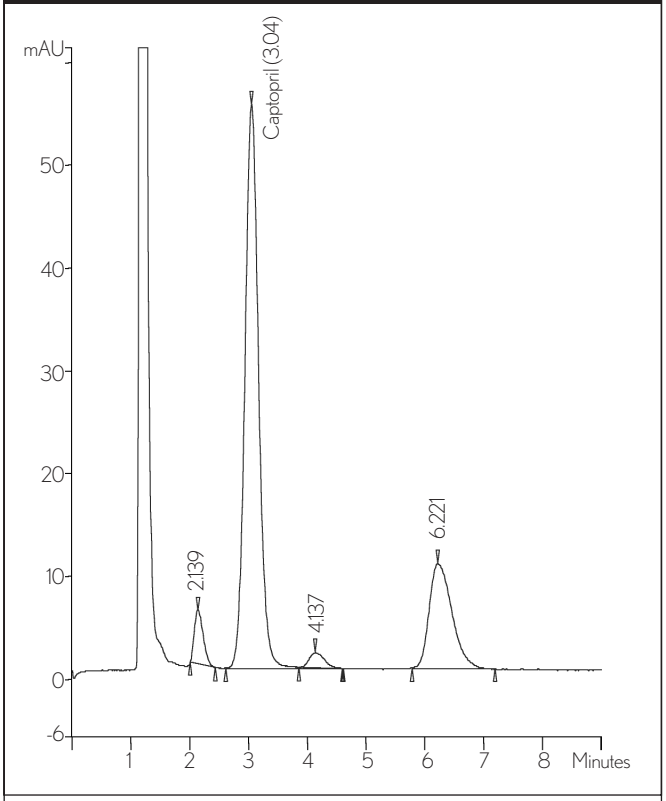
Following the results of the pilot study, formulation No. 1 was selected as the most promising one. Three identical batches were prepared separately and placed in 50 mL brown Veral flasks. The three batches of 1 mg/mL captopril were stored according to ICH (International Conference on Harmonisation) temperature recommendations [29] (i.e. at room temperature, at 5 ± 3°C, and at 40 ± 2°C). In contrast, humidity was not monitored because all the solutions were stored in well-closed glass bottles. Three flasks per batch were sampled at each time point and for each condition of storage; captopril content and pH were measured in triplicate in each sample, and visual and olfactory examinations were carried out. The microbiological tests were performed at time 0 and after six and 24 months of storage. Batches with losses in drug concentration smaller than 10% were defined as stable.

Results and discussion

HPLC method

The chromatograms obtained after forced degradation by heat, acid, base, oxidising agent and daylight showed an excellent separation between the peak of captopril (retention time three minutes) and those of the breakdown products (Figure 2). For example, H_2O_2 degradation produced two peaks (retention times 4.1 and 6.3 minutes) completely separated from that of captopril. Under these stress conditions there was a distinct peak separation between the drug and its degradation products as shown by the standard deviation (SD) of the absorbance spectra. In all cases, the SD of the average purity parameter of all peaks was lower than 1nm (for example: SD = 0.344 for H_2O_2 degradation). Moreover, in the best and worst correlations, the similarity factor was 0.9995 and 0.996 respectively, and the dissimilarity factor was 0.031 and 0.087 respectively.

Figure 2: High-performance liquid chromatogram obtained after one-hour degradation of a captopril solution in the presence of H_2O_2 at room temperature



Retention times are: captopril 3.0 minutes, degradation products 4.1 and 6.3 minutes

Analytical validation data

The linearity coefficient of determination was r^2 0.999 and the relative standard deviation (percentage of RSD) of response factor (peak area/concentration) was < 2%. The relative standard deviation (percentage of RSD) of intraday and interday variation was 1.58 and 2.02%, respectively.

Pilot stability study

The 10 formulations examined in the month-long pilot study and the stability results are shown in Table 1. After one month, no breakdown product was seen in the control formulation (No.1, captopril 1 mg/mL and EDTA-Na 0.1% in water). The stability of captopril in the nine other formulations already described in the literature was consistent with published results. These results showed that captopril solutions prepared by dissolving commercial tablets [19-21, 24] had a limited stability despite the addition of ascorbate and/or EDTA-

Table 1: Pilot study of 1 mg/mL captopril solutions stored for one month in 10 mL brown flasks

Ingredients	Formulation									
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 7	No. 8	No. 9	No. 10
Source of captopril:										
Powder	+	+	+	+	+	-	-	-	-	-
Tablets	-	-	-	-	-	+	+	+	+	+
Solvent:										
Purified water	+	+	+	-	+	+	+	+	-	+
Ora-Plus / Ora-Sweet (1:1)	-	-	-	+	-	-	-	-	+	-
Antioxidant:										
EDTA-Na (1 mg/mL)	+	-	+	-	-	+	-	+	-	-
Ascorbate (5 mg/mL)	-	+	+	-	-	-	+	+	-	-
pH at time 0	3.35	2.82	3.07	3.92	3.08	3.32	2.89	3.04	3.94	3.09
pH after one month:										
At room temperature	3.26	2.72	3.01	3.86	3.00	3.32	2.72	3.03	3.97	3.08
At 5 ± 3°C	3.33	2.91	2.97	3.92	2.91	3.36	2.74	2.73	3.85	3.05
% captopril remaining after one month (± SD, n = 3):										
At room temperature	100.3 ± 0.55	86.0 ± 1.00	93.0 ± 0.40	64.7 ± 2.62	94.7 ±4.82	94.9 ± 5.55	89.9 ± 0.10	92.4 ± 0.65	71.5 ± 1.20	93.6 ± 1.48
At 5 ± 3°C	99.7 ± 1.10	92.3 ± 0.58	96.7 ± 1.15	85.8 ± 3.65	95.1 ± 2.56	97.8 ± 3.09	93.6 ± 0.58	100.0 ± 1.03	81.9 ± 2.27	97.0 ± 0.98

Na. This may be explained by the presence in the tablets of enough metal ions to catalyse oxidation. The solutions prepared with Ora-Sweet and Ora-Plus also showed a limited stability with formation of a yellow colour within three weeks.

Long-term study

Based on the results of the pilot study, formulation No. 1 was selected and examined further in a long-term stability study. Organoleptic observations (visual and olfactory) did not reveal any noticeable change over the entire storage time. The pH in all preparations (3.33 ± 0.01) remained constant throughout. This value is considered to be in the optimal range for captopril conservation [9, 19]. Furthermore, slightly acidic pH values such as these are well tolerated orally. For example, the Ora-Sweet and Ora-Plus preparations have pH values in the range 4.0 to 4.5.

Captopril concentrations remained remarkably stable over the entire study at 5°C and at room temperature (Table 2). At 40°C, a marginal drop was seen such that the concentration had decreased by a few per cent to 95.8 ± 0.68% after 12 months.

No microbial growth (aerobic, anaerobic or fungal) was detected during the study in any of the samples stored at 5°C, room temperature and 40°C. A number of factors may explain this favourable outcome, namely:
1. The microbiological purity of the water used
2. The acidity of the solution
3. The known bacteriostatic effect of EDTA-Na [30, 31]

Table 2: Long-term stability study of formulation No. 1

Time	Storage temperature		
	5 ± 3°C	22 ± 2°C [a] (room temperature)	40 ± 2°C
	Percentage of initial concentration [b] remaining ± SD, n = 9 (lower and upper 95% confidence limits)		
Day 0	100.0 ± 0.94 (99.29-100.71)	100.0 ± 0.94 (99.29-100.71)	100.0 ± 0.94 (99.29-100.71)
7 days	100.6 ± 0.87 (99.94-101.26)	101.4 ± 0.73 (100.74-102.06)	100.7 ± 0.76 (100.13-101.27)
14 days	100.1 ± 0.56 (99.68-100.52)	98.9 ± 0.80 (98.30-99.50)	100.3 ± 0.32 (100.06-100.54)
21 days	98.0 ± 0.77 (97.42-98.58)	97.6 ± 0.64 (97.12-98.08)	97.8 ± 0.62 (97.33-98.27)
1 month	97.5 ± 1.20 (96.60-98.40)	97.9 ± 0.61 (97.44-98.36)	96.2 ± 0.85 (95.56-96.84)
2 months	103.4 ± 1.02 (102.63-104.17)	102.6 ± 0.61 (102.14-103.06)	102.6 ± 0.74 (102.04-103.16)
3 months	98.0 ± 0.32 (97.76-98.24)	98.8 ± 0.33 (98.55-99.05)	98.3 ± 0.16 (98.18-98.42)
6 months	98.5 ± 0.22 (98.33-98.67)	99.3 ± 0.37 (99.02-99.58)	100.1 ± 0.15 (99.99-100.21)
8 months	100.4 ± 2.50 (98.51-102.29)	101.4 ± 0.72 (100.86-101.94)	96.5 ± 1.53 (95.35-97.65)
12 months	102.7 ± 2.47 (100.84-104.56)	103.2 ± 0.92 (102.51-103.89)	95.8 ± 0.68 (95.29-96.31)
18 months	104.6 ± 0.32 (104.36-104.84)	103.6 ± 0.86 (102.95-104.25)	ND
24 months	101.4 ± 0.56 (100.98-101.82)	100.6 ± 0.76 (100.03-101.17)	ND

Key: [a]: ICH recommendation is 25 ± 2°C; [b]: initial concentration: 1.01 (± 0.01) mg/mL; ND = not determined

These results demonstrate that captopril can be prepared in oral solutions stable for two years or more when stored at room temperature or at 5°C. Three characteristics of the proposed formulation are worth mentioning:

1. It appears important to use captopril in powder form rather than dissolved from tablets liable to liberate compounds (e.g. metal ions) that accelerate the oxidation of the drug in solution
2. The solvent used was purified water containing neither microbes nor metal ions
3. The addition of EDTA-Na inactivates traces of metal ions released by the container

When comparing various captopril solutions, Lye et al. [20] found that EDTA-Na 0.1% and methylcellulose 2% afforded a good stability at one month. We did not add methylcellulose to our preparations because preliminary tests discounted its value and even suggested the possible liberation of traces of metal ions [20].

The duration of stability was determined with solutions kept in well-closed glass bottles that were opened 11 times to remove samples. In clinical practice, the duration of stability has been set at one month after the first opening, as based on systematic tests (data not shown) in which the flasks were opened six days per week for one month.

Safety of EDTA in paediatric patients

EDTA-Na is widely used in topical, oral and parenteral pharmaceutical formulations. It is also extensively used in cosmetics and foods. The usual concentrations employed in pharmaceutical formulations are in the range 0.005-0.1% w/v [32]. To the best of our knowledge, no data or guidelines have been published regarding accepted doses in paediatric patients. The WHO (World Health Organization) has set an estimated acceptable daily intake for disodium edetate in foodstuffs at up to 2.5 mg/kg body-weight [33].

Our 1 mg/mL captopril solution is stabilised with 1 mg/mL EDTA-Na.The usual titrate dose of captopril administered to premature neonates and newborns being 0.5 mg/kg/dose, young patients administered captopril 0.5 mg/kg/dose given every six to 24 hours will receive up to a maximum of 2 mg EDTA-Na per day, which is below the WHO limit of 2.5 mg/day tolerated in foods. It should also be noted that for infants who weigh 12 kg or more, administration of marketed captopril preparations (usually dosed at 12.5, 25.0 and 50.0 mg) is recommended.

Conclusion

The liquid formulation developed and validated at our hospital offers an outstanding chemical and microbiological stability (at least two years at room temperature). This can be explained by using purified water as solvent and mastering the major factors favouring captopril oxidation. The determining role of EDTA-Na is worth stressing, because it combines cation-chelating and bacteriostatic properties. In summary, we propose an oral formulation of captopril for paediatric use which can be prepared with ease by qualified professionals, is stable for at least two years at room temperature, and allows individualised dosage and easy administration, even to newborn patients.

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