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Reduced bioavailability of atenolol in man: the role of bile acids

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Summary

The effect of bile acids on atenolol bioavailability was assessed using a randomised two-way cross-over study in eight healthy volunteers. Single dose kinetics were studied in fasted subjects using either Tenormin® or a preparation of atenolol containing bile acids. The co-administration of bile acids with atenolol resulted in a 30% reduction in AUC and a 28% reduction in C_{max} compared to Tenormin®. No significant differences in t_{max} or $t_{1/2}$ were detected. The potential clinical implications of the interactions between atenolol and bile acids in vivo are discussed, together with a possible explanation for the observed effects of food on atenolol bioavailability.

Introduction

Atenolol is a well studied and proven cardioselective β-adrenoceptor blocking drug, without membrane stabilising properties or partial receptor agonist activity, used in the treatment of hypertension and angina. Unlike the lipophilic β-blockers propranolol and metoprolol, atenolol does not undergo extensive hepatic first-pass metabolism when administered orally (Kirch and Gorg, 1982; Ledermann et al., 1985). The hydrophilic nature of atenolol reduces the likelihood of hepatic clearance while ensuring that the drug is mainly excreted intact by the kidneys (Brown et al., 1976; Fitzgerald et al., 1978). This property also results in a predictable pharmacokinetic profile with minimal intra- and inter-subject variation (Lederman et al., 1985).

Although subject to little metabolic conversion, only about 50% of orally administered atenolol enters the systemic circulation (Vergin and Nitsche, 1989). This probably results from poor absorption from the gastrointestinal tract (Brown et al., 1976; Conway et al., 1976; Mason et al., 1978; Yamaguchi et al., 1986a). Melander et al. (1979) showed that food intake could further impair the absorption of atenolol from oral

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formulations of the drug, however, similar effects were not observed with lipophilic β-blockers (Melander et al., 1977).

Interestingly, a number of β-blockers have been shown to have surface active properties and also found to be capable of self-association (Attwood and Agarwal, 1979). Other studies have shown that hydrophilic β-blockers such as atenolol and nadolol strongly interact with bile acid micelles in vitro (Yamaguchi et al., 1986b,c). Previous investigations have demonstrated that bile acids are capable of both increasing (Kakemi et al., 1970; Kimura et al., 1971, 1972; Miyazaki et al., 1980; Cole et al., 1992) and decreasing (Yamaguchi et al., 1986b,c) the absorption of drugs from the gastrointestinal tract. In humans, the presence of a functioning gall-bladder ensures that after a prolonged fast the gastrointestinal tract is largely free from bile acids, however, subsequent consumption of food results in the release of bile acids into the duodenum. The effect of gall-bladder emptying on drug bioavailability has been discussed by Cole et al. (1992).

The present study investigates the possibility that the co-administration of atenolol with bile acids reduces atenolol bioavailability by impeding absorption of the drug from the gastrointestinal tract of fasted subjects. The possible clinical implications are discussed.

Materials and Methods

Atenolol B.P. grade was obtained from Becpharm Ltd (U.K.) for formulation and bioavailability studies and from Sigma (U.K.) as an analytical standard. Bile acids (composition as described in White et al. (1991)) and sucrose (B.P. grade) were obtained from Consolidated Chemicals Ltd (Wrexham, U.K.). Hydroxypropylmethyl cellulose phthalate HP55 (HPMC-phthalate), povidone, diethyl phthalate, B.P. or USP.NF grade, were supplied by Stancourt, Sons & Muir Ltd. Hard gelatin capsules, Licaps®, white opaque size 1, were purchased from Capsugel Ltd (Pontypool, U.K.). All other chemicals and solvents used were of an appropriate grade and obtained from Sigma, BDH or Metlab Ltd (Hawarden, U.K.). Tenormin® (Stuart Pharmaceuticals) 50 mg tablets were obtained from commercial sources.

Manufacture of dosage forms

The manufacturing procedure used was similar to that described by Cole et al. (1992). The bile acid/atenolol formulations consisted of sugar spheres coated with a mixture of atenolol and bile acids, subsequently enteric coated and packed into size 1 hard gelatin capsules. The drug coating solution was made by dissolving povidone and atenolol into an alcoholic solution containing bile acids. The weight ratio of bile acids to atenolol was approx. 2:1. This solution was bottom sprayed on to sucrose using a Uni-Glatt® fluidised bed (air suspension system). The resulting spheres were then coated, using the same system, with an enteric coating solution containing HPMC phthalate, diethyl phthalate and water in ethanol. In all cases the temperature and air flow in the Uni-Glatt® system were sufficient to rapidly evaporate the solvent(s) used. The final formulations, before and after enteric coating, were assayed for atenolol content by the HPLC method described below and the capsule-sphere fill weight determined accordingly. Capsules were manufactured to contain 50 mg of atenolol.

High performance liquid chromatography of the bile acid / atenolol formulation

A potency and stability-indicating reverse-phase HPLC assay for atenolol was developed from methods previously described by Verghese et al. (1983) and Harrison et al. (1985). The system consisted of a Varian® 5500 liquid chromatography system incorporating a programmable ultraviolet/visible detector, a 250 mm × 4.6 mm (i.d.) S5 CN bonded nitrile Spherisorb® HPLC column (Phase Separations Ltd, U.K.) and a 50 mm × 4.6 mm (i.d.) guard column constructed of the same material. The mobile phase was 0.05 M potassium phosphate buffer, pH 7.0:acetonitrile (9:1) with a flow rate of 1.5 ml min^{-1}. Detection was at 224 nm and the detector sensitivity was controlled by a Varian® 4270 integrator set to an attenuation of 32. The total sample volume injected was 10 μl. The

approximate retention time for atenolol was 9 min. The linear response range for atenolol was 1.0–5.0 mg ml^{-1} with a correlation coefficient of 0.925. Atenolol standards were made up in methanol:water (9:1). System suitability tests, using six injections of standards, indicated that the performance of the system remained within a 2% confidence limit based on coefficient of variance. To analyse atenolol in bile acid/atenolol formulations, spheres were weighed, dissolved in methanol:water (9:1), sonicated (Decon Ultrasonics Ltd, U.K.) until dissolved and filtered through a 0.45 μm cellulose acetate filter before HPLC analysis in duplicate or triplicate. The other components of the formulation, bile acids, povidone, HPMC phthalate, diethyl phthalate and sucrose did not interfere with the detection of atenolol. Determination of atenolol in human plasma was carried out by the HPLC method described by Harrison et al. (1985).

Dissolution and stability testing

Dissolution testing of the bile acid/atenolol formulation was carried out using the method of Cole et al. (1992). Bile acid/atenolol formulations used for clinical studies were stored at room temperature in the dark for 3 months following manufacture. All clinical studies were performed within 6 weeks of manufacture.

Clinical studies

Eight healthy subjects, two female and six male, aged between 26 and 46 years, participated in the study and were employees or students of the Academy of Medicine, Bialystok, Poland. The average height and weight of the subjects was 171 ± 11 cm (S.D.) and 70.6 ± 12.5 kg (S.D.) respectively. Subjects were shown to be in good health by a physical examination and a series of hospital laboratory tests. To comply with Polish ethical approval requirements the protocol for the study was approved by an independent panel of researchers at the Academy of Medicine, Bialystok and the subjects gave their informed consent. The subjects were asked to abstain from taking any medication for 2 weeks before the start of the study and until after the collection of the last blood sample. Alcohol, tea, coffee and other xanthine-containing beverages were prohibited for the 24 h period before the study until its completion. Food was withdrawn for 12 h over the night preceding each part of the study. A light breakfast was allowed 3 h post dose after which time the subjects were allowed to follow their normal daily diets. The study was a randomised two-way cross-over design with the subjects receiving either Tenormin® or the bile acid/atenolol formulation containing an equivalent dose of 50 mg of atenolol. The medication was taken with approx. 250 ml of boiled tap water. The zero time blood samples were taken within a 5 min period preceding the administration of the medication. Subsequent samples were taken at 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4, 6, 8, 10, 12 and 24 h. Blood samples were collected into lithium heparin tubes, gently mixed and centrifuged at 2.0×10^4 $g \cdot \min$ within 15 min of collection. Separated plasma was transferred into clean tubes and stored at -20°C.

After a 1 week interval the subjects received the alternative medication and the blood sampling protocol repeated. Plasma samples were analysed by the HPLC method described above. No degradation of atenolol was detected during the period of storage at -20°C.

The results of the clinical study were evaluated using the observed values of maximum plasma concentration C_{max} and t_{max}. The area under the plasma concentration curves was calculated using the trapezoidal rule, and the plasma half-life determined. The statistical significance of the results was assessed by paired t-test for the continuous variables AUC, C_{max} and $t_{\frac{1}{2}}$. The discrete variable, t_{max}, was evaluated using the Wilcoxon paired-sample test.

Results

The average plasma concentrations of atenolol after the administration of Tenormin® and the bile acid/atenolol formulation are shown in Table 1. These results indicate that mean plasma levels of atenolol resulting from the administration of the bile acid/atenolol formulation remain consistently below those observed for Tenormin®.

Table 2 lists the pharmacokinetic parameters for each of the atenolol formulations. The AUC for the bile acid/atenolol formulation was more than 30% lower ($P = 0.016$) than that observed for Tenormin® (1455 compared to 2324 ng ml^{-1} h). A reduction in C_{max} of 28% ($P = 0.037$) using the bile acid/atenolol formulation was also observed. C_{max} values were 165 and 230 ng ml^{-1} for the bile acid/atenolol formulation and Tenormin®, respectively. No significant difference in $t_{\frac{1}{2}}$ was detected between the two atenolol formulations, however, the differences in t_{max} approach significance suggesting slightly slower uptake (see Table 2). This observation is probably explained by the dissolution characteristics of the bile acid/atenolol formulation. Dissolution studies showed that atenolol was not released in detectable amounts from the enteric-coated bile acid/atenolol formulation at pH 1.2, whereas, release of atenolol from the bile acid/atenolol formulation at pH 7.4 was found to be rapid and complete and therefore showed the performance of the formulation in vitro to be similar to those described by Cole et al. (1992). The slight delay in t_{max} with the bile acid/atenolol formulation may therefore be due to the time taken for the enteric-coat to be removed from the delivery vehicle spheres when passing from the acid environment of the stomach into the duodenum. Disintegration and dissolution of Tenormin® would begin in the stomach and be rapidly completed in the duodenum. Dissolution characteristics and potency of the bile acid/atenolol formulation did not significantly alter during the period of the clinical study.

TABLE 1

Summary of plasma atenolol concentrations in subjects receiving the bile acid / atenolol formulation and Tenormin®

Time (h)	Bile acid/atenolol formulation (ng ml^{-1})	Tenormin® (ng ml^{-1})
0	0	0
0.5	11 ± 20	75 ± 46
1.0	78 ± 39	169 ± 74
1.5	100 ± 41	178 ± 64
2.0	110 ± 44	183 ± 65
2.5	130 ± 56	186 ± 58
3.0	144 ± 50	187 ± 75
3.5	124 ± 45	200 ± 85
4.0	112 ± 43	177 ± 67
6.0	92 ± 39	153 ± 60
8.0	70 ± 32	117 ± 41
10.0	58 ± 29	79 ± 30
12.0	44 ± 16	59 ± 20
24.0	15 ± 8	26 ± 15

Values are means (±S.D.) of the plasma atenolol levels in eight subjects. Individual values are in turn the means of duplicate or triplicate determinations.

TABLE 2

Pharmacokinetic data for subjects receiving the bile acid / atenolol formulation and Tenormin®

Subject	C_{max} (ng ml^{-1})	t_{max} (h)	$t_{1/2}$ (h)	AUC (ng ml^{-1} h)	AUC ratio
Tenormin®					
1	188	1.0	6.4	1667	
2	209	1.0	11.1	3275	
3	264	2.0	3.1	2198	
4	188	2.5	6.4	1568	
5	327	3.5	8.1	3203	
6	116	2.0	9.2	1226	
7	270	3.0	4.8	2143	
8	276	1.5	7.4	3308	
Mean	230	2.1	7.1	2324	
S.D.	67	0.9	2.5	837	
Bile acid/ atenolol					
1	247	3.0	5.1	1116	0.67
2	124	3.0	11.8	1120	0.34
3	178	1.5	5.2	2051	0.93
4	162	3.0	10.1	1727	1.10
5	157	3.5	6.2	1652	0.52
6	78	3.0	15.1	702	0.57
7	236	2.5	5.8	1413	0.66
8	141	3.5	8.9	1859	0.56
Mean	165	2.9	8.5	1455	
S.D.	56	0.6	3.6	451	
P	0.017	0.091	0.146	0.016	

Discussion

The present study shows that the co-administration of bile acids with atenolol results in about a 30% reduction in drug bioavailability in human volunteers compared to Tenormin®. This difference in bioavailability could not be explained by

poor dissolution characteristics of the bile acid/atenolol formulation or degradation of atenolol. Only C_{max} and AUC were affected with no significant differences observed in t_{max} and $t_{\frac{1}{2}}$. Previous studies have shown that the rate and extent of absorption of β-blockers from the gastrointestinal tract is determined by their relative hydrophobicity (Yamaguchi et al., 1986a–c). Hydrophobic β-blockers, such as propranolol and metoprolol, are rapidly and efficiently absorbed in vivo, whereas this is not the case for hydrophilic β-blockers, for example, atenolol and nadolol. Of further interest is the tendency of hydrophilic β-blockers to become tightly associated with bile acid micelles, a phenomenon which has been suggested as an explanation for their reduced absorption from the gastrointestinal tract in vivo (Yamaguchi et al., 1986b,c). In contrast, bile acids have been shown to increase the bioavailability of lipophilic β-blockers (Gasco et al., 1984), however, they may also cause distinct changes in the pathways by which they are absorbed (White et al., 1991).

It is well known that the consumption of food results in the contraction of the gall-bladder and the subsequent release of bile acids into the duodenum in humans. Therefore, in studies designed to evaluate the effects of food on drug bioavailability, it is inevitable that the concomitant effects of bile acids upon drug absorption are also observed. Studies of this type using atenolol show a considerable decrease in drug bioavailability with food intake (Melander et al., 1979). Interestingly, the results of the present study show an even more extensive reduction in atenolol bioavailability with bile acids than in the presence of food. When taken together with the in vitro findings concerning the interaction of hydrophilic β-blockers and bile acids (Yamaguchi et al., 1986b,c), these results suggest that it is the presence of bile acids in the duodenum, released by the gall-bladder in response to food, which reduces atenolol bioavailability and not the food per se. Further support for this hypothesis follows the observation that (i) food has little or even an enhancing effect upon the bioavailability of hydrophobic β-blockers (Melander et al., 1977) and (ii) the micellar interaction between lipophilic β-blockers and bile acids has been shown to be much weaker than bile acids and hydrophilic β-blockers (Yamaguchi et al., 1986b,c).

The results of the present study may be of considerable clinical significance given that bile acids are used for the dissolution of gall-stones (Bachrach and Hofmann, 1982; Maton et al., 1982). Patients on concomitant atenolol therapy may experience a reduction in the effectiveness of their treatment. Furthermore, bile acids have also been used as a marker for hepatic portal blood flow in cirrhosis of the liver (Testa et al., 1988, 1989; Ghia et al., 1990). Atenolol, and other β-blockers, are also used in the treatment of elevated hepatic portal blood pressure in cirrhosis of the liver (Hillon et al., 1982; Kroeger and Groszmann, 1985; Groszmann, 1987; Reichen and Lebrec, 1987). Interaction between bile acids and atenolol in subjects with cirrhosis of the liver could result in the failure of the management of this condition.

In conclusion, this study demonstrates that bile acids reduce the bioavailability of the hydrophilic β-blocker atenolol to a similar or greater extent than previously observed in the presence of food. Given the nature of the physico-chemical interactions between hydrophilic β-blockers and bile acids it is likely that the effects of food on atenolol bioavailability results from the presence of bile acids. The effect of bile acids on atenolol bioavailability may have important implications for the clinical use of both bile acids and atenolol.

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