Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Controlled Release 95 (2004) 515–520
journal of controlled release
www.elsevier.com/locate/jconrel

Evaluation of novel starch acetate–diltiazem controlled release tablets in healthy human volunteers

Ossi Korhonen*, Harri Kanerva, Mika Vidgren, Arto Urtti, Jarkko Ketolainen

Department of Pharmaceutics, University of Kuopio, P.O. Box 1627, FIN-70211 Kuopio, Finland

Received 24 June 2003; accepted 23 December 2003

Abstract

Highly substituted starch acetate can be used to control drug release from directly compressed tablets in vitro. The aim of this study was to evaluate controlled release properties of starch acetate in vivo in humans. Three starch acetate tablet formulations with different in vitro release rates for diltiazem (fast, moderate and slow) were developed. An open, single dose, randomised, four treatment, four period, four sequence cross-over pharmacokinetic study was conducted in eight healthy volunteers. Diltiazem concentrations in plasma were determined by HPLC. Concentration–time profiles of the formulations differed: mean C_{max} and $AUC_{0-\infty}$ values of the fast, moderate and slow formulations were 95, 69, 31 ng/ml and 610, 511, 231 ng h/ml, respectively. In vitro–in vivo correlation (IVIVC) was analysed according to the cumulative area under the curves and in vitro release profiles. Acceptable limits of prediction errors were achieved for C_{max} and $AUC_{0-24\ h}$. The moderate formulation and commercial reference tablet showed similar in vitro release profiles and diltiazem concentrations in plasma. In conclusion, direct compression starch acetate formulations control drug release in humans.

Keywords: Starch acetate; Matrix tablet; Controlled drug release; Pharmacokinetics; IVIVC

1. Introduction

Highly substituted starch acetate was recently introduced as a matrix-forming excipient for the direct compression of pharmaceutical tablets [1–4]. Drug release from the tablets could be easily modified over a wide range for various drugs. The mechanism of drug release from the starch acetate matrix tablets appears to be diffusional in most cases. The most significant variables that affect the release of drugs from the starch acetate matrix are the particle and powder properties of starch acetate, tablet properties (i.e. porosity), the ratio of drug and starch acetate concentrations in the formulation, and the physicochemical nature of the drug. The drug release profile can be governed by adjusting these factors. The potential functionality of starch acetate as a pharmaceutical excipient, however, has not been tested in vivo.

An excipient that modifies drug release in vitro should also be able to regulate drug release in vivo, and prolong drug concentrations within the therapeutic window. Ideally, in vitro and in vivo performance

* Corresponding author. Tel.: +358-17-16-2458; fax: +358-17-16-2252.
E-mail address: Ossi.Korhonen@uku.fi (O. Korhonen).

0168-3659/$ - see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.jconrel.2003.12.026

should correlate with each other. If the in vitro–in vivo correlation (IVIVC) is established, an in vitro dissolution test may be indicative of the in vivo performance (e.g. in the case of bioequivalence). Different correlation levels, acceptance limits and predictability of IVIVC have been introduced in FDA industrial guidelines as "extended release oral dosage forms: development, evaluation, and application of in vitro/in vivo correlation" [5]. The most common process for developing a Level A IVIVC is to (1) develop formulations with different release rates (fast, moderate and slow), (2) obtain in vitro dissolution profiles and in vivo plasma concentration profiles for these formulations and (3) estimate in vivo absorption profiles by an appropriate deconvolution technique for each formulation and subject.

The aim of this study was to evaluate if starch acetate is able to control drug release from tablets in humans. The calcium channel blocker, diltiazem, was used as a model drug because it is widely used as sustained release tablets in clinical practice. Predictive IVIVC model was developed for the starch acetate–diltiazem tablets, and performance of the experimental formulations was compared to an established clinical formulation.

2. Materials and methods

2.1. Materials

The matrix-forming excipient, starch acetate degree of substitution 2.7, was manufactured by VTT (Department of Chemical Technology, Materials Technology, Finland). A particle size fraction <500 μm was used. Magnesium stearate (Ph.Eur. grade) was used as a lubricant in tableting. Diltiazem HCl (Ph.Eur. grade, Orion Pharma, Finland), hereafter referred to as diltiazem, was used as supplied. Dilzem depottabl 90 mg (Orion Pharma) was used as a reference product.

2.2. Formulations

Three different starch acetate–diltiazem tablets were formulated to produce fast, moderate and slow drug release profiles. These formulations contained 50% (fast, D50), 35% (moderate, D35) and 15% (slow, D15) of diltiazem. Each formulation contained 90 mg of diltiazem and 0.5% (m/m) of magnesium stearate.

2.3. In vitro dissolution

In vitro dissolution experiments were carried out using USP XXVI (paddle apparatus), operating at 100 rpm. The dissolution medium was 900 ml of pH 4.2 acetate buffer, maintained at $+37$ °C (USP XXVI monograph for diltiazem hydrochloride extended-release capsules). Acetate buffer pH 4.2 was used as a dissolution medium to ensure the chemical stability of diltiazem during experiment. Samples of 5 ml were collected at 15 and 30 min, and then at 1, 2, 3, 4, 6, 8, 10 h. The samples were replaced by equivalent volume of dissolution medium. Three tablets of each formulation were used in the dissolution test.

Diltiazem was quantitated using HPLC with UV detection. The column was packed with a reverse phase C-18 support (Inertsil ODS-3, 5 μm, 150×4.0 mm; GL Sciences, Japan) operating at $+40$ °C. The injection volume was 20 μl, the mobile phase was acetonitrile–water with trifluoroacetic acid (30:70:0.03), its flow rate was 1.0 ml/min, and the wavelength of detector was 237 nm. The retention time of diltiazem was 4.2 min. A linear correlation ($r^2>0.999$) was obtained between the peak areas and diltiazem concentrations over the range of 1–100 μg/ml. The coefficient of variation for intra-day precision was 0.27% at 25 μg/ml, and the bias% of intra-day accuracy was -0.72% at 25 μg/ml.

2.4. In vivo study and demographics

Eight volunteers (three males and five females) completed the study. The mean age of the volunteers was 22.6 years (S.D. 3.11; range 19–28 years), mean weight 65.5 kg (S.D. 14.6; 51–91 kg) and mean height 173.5 cm (S.D. 10.8; 162–188 cm). Body mass index of all volunteers was included in the range 19–26.

An open, single dose, randomised, four treatment, four period, four sequence cross-over trial was conducted in eight healthy volunteers to evaluate concentrations in plasma after ingestion of three formulations and the reference product. Each treat-

ment period was followed by a 2-week wash out period to eliminate the effect of each tested dose before the next treatment. C_{max} is the maximum drug concentration in plasma, and it was directly recorded from the concentration profiles. $AUC_{0-\infty}$ is an area under the drug concentration in plasma versus time curve. It was calculated using trapezoidal rule by using the WinNonlin Professional software (version 4.0.1, Pharsight, USA).

The present study was designed according to ethical and regulatory requirements for the pharmacokinetics trial. The study was approved by the Ethics Review Committee on Human Research at the University of Tartu, Estonia and the State Agency of Medicines, Tartu, Estonia.

2.5. Assay of diltiazem in plasma samples and validation of the analysis method

The recovered plasma samples were frozen at −20 °C immediately after separation. After thawing plasma aliquots (700 μl) were mixed 7 μg/ml verapamil (internal standard) solution. Then, acetonitrile (900 μl) was added to each sample, centrifuged and kept frozen overnight at −20 °C to facilitate the separation of the aqueous and organic phases. From frozen samples 600 μl of the organic phase was evaporated to dryness, reconstituted in the mobile phase, vortexed and centrifuged. Aliquots were transferred to an auto sampler vial. The HPLC analysis was done as follows: a column (Lichrosorb RP-18, 5 μm, 250 × 3.2 mm; Alltech Associates, USA), an injection loop of 40 μl, mobile phase (acetonitrile–0.01 M phosphate solution (Na_2HPO_4)–triethylamine, 15:85:1), flow rate of 0.7 ml/min, UV detector wavelength of 237 nm, column temperature of 21–23 °C, temperature in autosampler 10 °C, and a run time of 13 min. All reagents were of analytical grade.

Analysis method was validated according to established international guidelines and requirements (Validation of Analytical Methods: Definitions and Terminology, ICH Topic Q2A, and Validation of Analytical Procedures: Methodology, ICH Topic Q2B). No interfering peaks were detected at the retention times of diltiazem (5.3 min) and verapamil (8.3 min). A linear correlation (r^2>0.999) was obtained between the ratio of peak area and diltiazem concentration between the range of 10–3000 ng/ml. The coefficient of variation of the slope was 7.2%. The limit of quantification (10 × background noise) was 10 ng/ml. Precision and accuracy of the method was evaluated at concentrations of 3000, 300 and 30 ng/ml. Precision of the method was assessed on the basis of the coefficient of variation in quality control samples and accuracy was calculated as the bias% of these samples. The coefficient of variation of intra- and inter-day precision was 2.0–5.6% and 2.9–6.7% at all concentrations, respectively. The bias% of intra- and inter-day accuracy was 0.7–2.3% and −0.5 to 0.9%. No decrease in the content of quality control samples was observed in the freezer or autosampler.

2.6. Establishment of in vitro–in vivo correlation (IVIVC)

The cumulative $AUC_{0-24\ h}$ of time profiles (AUCcum(t)) was generated from averaged concentration–time profiles of diltiazem in plasma without further time scaling. AUCcum(t) of each formulation was fitted to the Hill equation (Eq. (1)) and constants a, b and c were determined [6]:

$$\mathrm{AUCcum}(t) = \frac{a \cdot t^b}{c^b + t^b} \quad (1)$$

where a, b and c are constants, and AUCcum(t) is the cumulative area under curve at time t.

Average profiles of in vitro dissolution were fitted by Power Law equation from 2 up to 75%:

$$\frac{M_t}{M_\infty} = k \cdot t^n \quad (2)$$

where M_t/M_∞ is the fractional (0–0.75) drug release at time t, k is drug release rate constant reflecting matrix structure, and n is an exponent, which was used to characterise the transport mechanism. Next, a correlation analysis was conducted to find correlations between Hill's constants and in vitro drug release rates and diffusional exponents. Finally, the predicted AUCcum(t) was calculated from the in vitro–in vivo regression equation and Hill function. The internal predictability of IVIVC was evaluated to ensure the predictive performance of the model, using a percent prediction error (%PE) calculation.

Table 1
In vitro release parameters of Power Law equation and fitted Hill parameters for AUCcum(t), and prediction error (%PE) values for AUC_{0-24} and C_{max}

	In vitro profiles		In vivo Hill's constants			Prediction error%	
	Rate constant (k)	Diffusional exponent (n)	a	b	c	C_{max}	$AUC_{0-24\ h}$
D50	57.27	0.42	620.1	1.94	5.43	− 19.3	− 1.7
D35	43.15	0.42	512.7	2.01	5.61	− 9.7	− 8.2
D15	18.98	0.43	197.4	2.02	5.62	− 5.4	− 0.4
Mean	–	0.43	–	1.99	5.55	− 11.4	− 3.4
Standard deviation	–	0.01	–	0.04	0.11	7.1	4.2

Predicted $AUC_{0-24\ h}$ and C_{max} were compared to the observed $AUC_{0-24\ h}$ and C_{max} using Eq. (3):

$$\%PE = \left[\frac{(Obs - Pred)}{Obs}\right] \cdot 100\% \quad (3)$$

3. Results and discussion

3.1. In vitro dissolution

Release of diltiazem was affected by the concentration of diltiazem and tablet porosity in the formulation. Compaction force and breaking strength of tablets were 5.7 ± 0.3 kN, 148 ± 14 N and 14.5 ± 0.3 kN, 213 ± 12 N and 20.3 ± 0.9 kN, 451 ± 14 N, for formulations D50, D35, and D15, respectively. The coefficients of variation within formulations for dissolution profiles were below 4.4% in each case. Power Law equation could nicely fit all profiles up to 75% (r^2>0.99). Drug release mechanism followed Fickian diffusion in each formulation (equal diffusional exponents n) (Table 1), which agreed with previous results [3]. As the amount of diltiazem and the porosity of tablet increased, so did the rate of drug release (Fig. 1). Since the diffusional exponents (n) were practically equal in all formulation, constant k can be used as drug release rate constants for different formulations (Table 1). According to Fig. 1, the profiles of formulation D35 and the reference product show that drug release from these formulations is superimposable. According to in vitro results, three diltiazem–starch acetate tablet formulations with different drug release rates (fast, moderate and slow), but equal drug release mechanisms were produced. Reproducible dissolution method was achieved, and an appropriate equation to describe dissolution profiles was used.

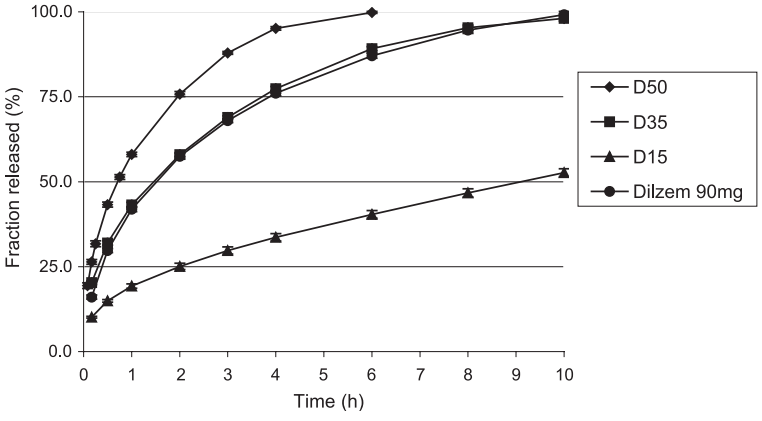

Fig. 1. In vitro mean dissolution profiles of diltiazem (90 mg) from starch acetate formulations (D15, D35, D50) and a commercial reference product (Dilzem 90 mg depottabl).

3.2. Bioavailability

Plasma profiles of diltiazem clearly differed between the three starch acetate formulations (Fig. 2 and Table 2). Same rank order exists between in vitro and in vivo profiles, and C_{max} and $AUC_{0-\infty}$ decreased as in vitro dissolution drug release rate decreased, which confirms that starch acetate is able to modify drug release from tablets in humans. In addition, in vivo profiles of D35 and reference product were almost superimposable, but the statistical bioequivalency was not achieved due to the large inter-subject variability (Fig. 2).

3.3. In vivo–in vitro correlation

Diltiazem follows at least a two-compartment model after intravenous administration [7]. Therefore, the Wagner–Nelson method cannot be used for the establishment of IVIVC. Deconvolution methods and the Loo–Riegelman method are commonly used to analyse IVIVC for two-compartment model drugs [8]. Since the reference administration (e.g. intravenous or an oral solution) is needed in these methods, an alternative method was used to establish IVIVC, without any reference administration.

Establishment of IVIVC was initiated by plotting the mean AUCcum(t) against time and fitting those curves to the Hill equation. Generated Hill equations fit well with the AUCcum(t) profiles (r^2>0.99), and each constant was statistically significant. Constants b and c were almost similar and independent of formulation, while constant a was strongly dependent on formulation (Table 1). After that, the mean values of

Table 2
Pharmacokinetic parameters (n = 8)*

	C_{max} (ng/ml)	$AUC_{0-\infty}$ (ng h/ml)
D50	95.4 (14.4)	609.5 (174.4)
D35	68.8 (23.5)	510.7 (189.8)
D15	31.1 (8.0)	231.4 (107.4)
Dilzem 90 mg depottabl	70.7 (18.0)	532.6 (195.0)

* Standard deviations in parenthesis.

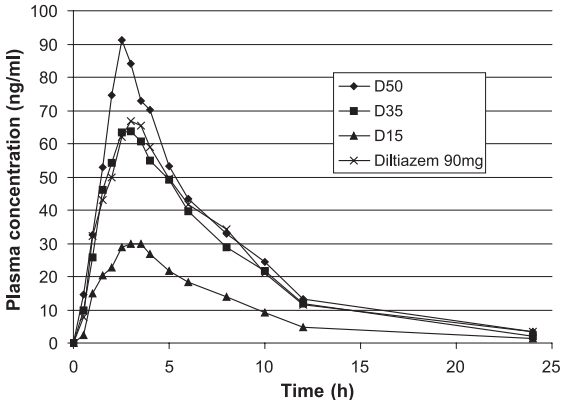

Fig. 2. Diltiazem plasma concentrations after per oral administration of starch acetate tablets (90 mg) (D15, D35, D50) and the reference commercial product (Dilzem 90 mg depottabl).

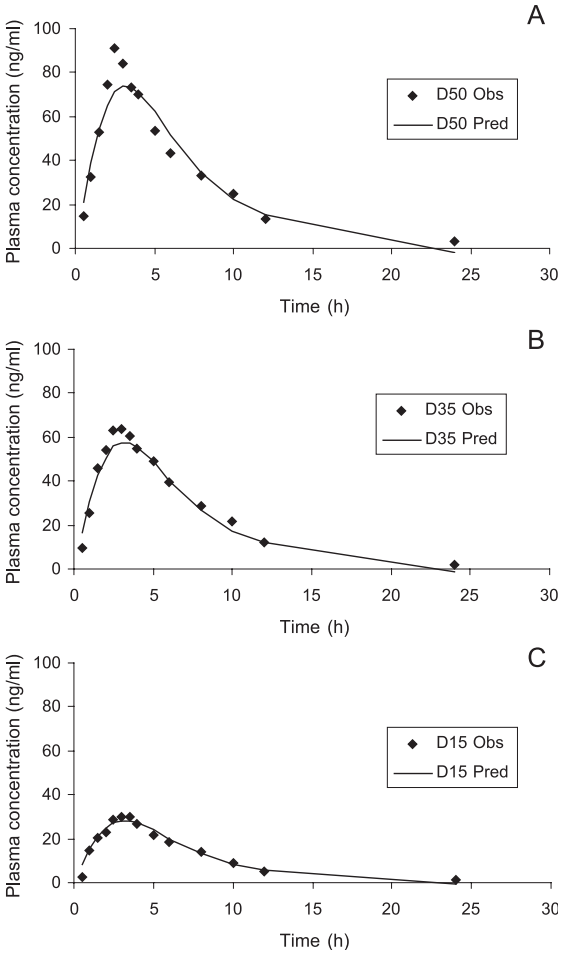

Fig. 3. Observed and predicted plasma concentrations from formulation D50, D35 and D15 in panels A, B and C, respectively.

constants b and c were used, and constant a was used to describe AUCcum(t) profiles (Eq. (4)).

$$\mathrm{AUCcum}(t) = \frac{a \cdot t^{2.0}}{5.6^{2.0} + t^{2.0}} \quad (4)$$

According to the correlation analysis, there was a strong positive correlation between in vitro drug release rate (k) and in vivo AUCcum(t), described by Hill's constant a, and the following linear regression equation was, therefore, obtained (Eq. (5)):

$$a = 10.4 \cdot k + 45.6 \qquad (r^2 = 0.993) \quad (5)$$

Evaluation of the internal predictability of IVIVC was based on calculations from the initial data sets (D15, D35 and D50). First, new Hill's constants a were determined using Eq. (5) and in vitro drug release rate constants (k) for each formulation. Next, predicted AUCcum(t) profiles were generated using Eq. (4) and respective new Hill's constants a. Finally, predicted plasma concentrations were rehabilitated from the predicted AUCcum(t) values (Fig. 3). Prediction errors on $AUC_{0-24\ h}$ and C_{max} for each formulation are presented in Table 2. All formulations reached the acceptance limits except the C_{max} of D50, which was outside of the 10% acceptance limit.

4. Conclusions

This is the first time when starch acetate based on direct compression matrix tablets were administered to humans in bioavailability study. In this study it was established that starch acetate is able to modify drug release with direct compressed tablets in humans. IVIVC was established without any reference administration of diltiazem, which follows multicompartmental pharmacokinetics, extensive first pass metabolism and large inter-subject variability. The combination of the cumulative AUC, the Hill equation and correlation and regression analyses enabled the establishment of a predictive IVIVC. Advantages of established IVIVC are direct determination of final plasma concentration (and cumulative AUC) of drug instead of indirect estimation of drug absorption, and simple mathematics. However, the predictive AUC_{0-24} and C_{max} values were slightly underestimated, and thus, it might be beneficial to test alternative mathematical equations for the description of profiles. In present study, it should be noted that in vitro drug release profiles followed same release mechanism, which simplified the generation of IVIVC. If the release mechanism changes between formulations, it might lead to more complicated relationships between in vitro and in vivo profiles, and difficulties to find reasonable parameters to describe in vitro and in vivo profiles.

Acknowledgements

The authors are grateful to Ms. Pirjo Hakkarainen for her skillful technical assistance. The financial support from TEKES (National Technology Agency of Finland) and Polymer Corex Ltd. are gratefully acknowledged. This study was also supported by grants from Eemil Aaltonen Foundation and the Pharmacal Research Foundation, Finland.

References

[1] O. Korhonen, P. Raatikainen, P. Harjunen, J. Nakari, E. Suihko, S. Peltonen, M. Vidgren, P. Paronen, Starch acetates—multifunctional direct compression excipients, Pharm. Res. 17 (2000) 1138–1143.

[2] O. Korhonen, S. Pohja, S. Peltonen, E. Suihko, M. Vidgren, P. Paronen, J. Ketolainen, Effects of physical properties for starch acetate powders on tableting, AAPS Pharm. Sci. Tech. 3 (4) (2002) Article 34 (http://www.aapspharmscitech.org).

[3] S. Pohja, E Suihko, M. Vidgren, P. Paronen, J. Ketolainen, Starch acetate as a tablet matrix for sustained drug release, J. Control. Rel. (in press).

[4] P. Raatikainen, O. Korhonen, S. Peltonen, P. Paronen, Acetylation enhances the tabletting properties of starch, Drug. Dev. Ind. Pharm. 28 (2002) 165–175.

[5] FDA guidance for industry: extended release oral dosage forms: development, evaluation, and application of in vitro/in vivo correlations. September 1997.

[6] G. Balan, P. Timmins, D.S. Greene, P.H. Marathe, In vitro–in vivo correlation (IVIVC) models for metformin after administration of modified-release (MR) oral dosage forms to healthy human volunteers, J. Pharm. Sci. 90 (2001) 1176–1185.

[7] P. Hoglund, L.G. Nilsson, Pharmacokinetics of diltiazem and its metabolites after single and multiple dosing in healthy volunteers, Ther. Drug Monit. 11 (1989) 558–566.

[8] Y. Qiu, E.E. Samara, G. Cao, In vitro–in vivo correlations in the development of solid oral controlled release dosage forms, in: D.L. Wise (Ed.), Handbook of Pharmaceutical Controlled Release Technology, vol. 1, Marcel Dekker, New York, 2000, pp. 527–549.