INVESTIGATION OF FENOFIBRATE DISSOLUTION RATE INCORPORATED ON SOLID DISPERSIONS INTO CHITOSAN

Bożena Grimling, Maria Szcześniak, Jan Meler, Janusz Pluta

Faculty of Pharmacy,
Department of Pharmaceutical Technology,
The „Silesian Piasts" memorial Medical University of Wroclaw
St. Borowska 211a, 50-556 Wroclaw, Poland
E-mail: bamag@interia.pl

Abstract
The aim of the study was to investigate the effect of chitosan on the dissolution of fenofibrate incorporated into this polymer carrier. The study investigated fenofibrate in physical mixtures at the drug to polymer ratio of 1:9, 3:7, and 5:5. The solubility investigation was performed by means of a dynamic method in a dissolution apparatus; mean amount of dissolved fenofibrate and the drug to polymer quantitative ratio in which the solid dispersion possessed the most beneficial properties improving the drug solubility were calculated. The study revealed a multi-fold increase (from 13 to 70 times) in fenofibrate solubility in the presence of chitosan, which increased with duration of the study and with increasing percentage of the polymer in formulations. The dissolution rates of fenofibrate in the presence of chitosan at the weight ratio 1:9 increased with the increment of the molecular weight of the chitosan. The obtained results may help develop new technologies for fenofibrate preparations with chitosan, with better solubility characteristics, and thus increased bioavailability of the drug.

Key words: *solid state, dissolution, fenofibrate, molecular weight of the chitosan.*

1. Introduction

Fenofibrate, a third generation fibric acid derivative, is a precursor, which is hydrolyzed by tissue and plasma esterases to the active metabolite fenofibric acid. Its pharmacological activity consists in reducing triglyceride and cholesterol concentration in plasma. Fenofibrate is a useful drug for the treatment of atherogenic dyslipidemias producing a substantial decrease in the levels of triglyceride-rich lipoproteins and an increase in high density lipoprotein cholesterol (HDL-C) levels [**1, 2**].

Solubility and permeability are the fundamental parameters controlling the rate and extent of drug absorption. Amidon and co-workers devised a Biopharmaceutics Classification System (BCS) that categorized drugs into four classes according to their solubility and permeability properties. The objective of the BCS is to predict in vivo pharmacokinetic performance of drug products from measurements of permeability and solubility. According to the BCS, fenofibrate is a Class II, low solubility and high permeability drug [**3, 4**].

Many methods are available to improve these characteristics, micronization, addition of solvent or surface-active agent. Solid dispersion is one of these methods, and involved a dispersion of drugs in an inert carrier in the solid state prepared by melting, solvent or solvent-melting method.

Solid dispersions with the use of polymers, especially chitosan, as a carrier, play an exceptional role. Chitosan, dispersing in water environment, causes a significant increase in the contact area of the drug with solution, increases its hydrophilic properties and may affect its crystalline structure. All these factors lead to increased solubility of the drug [**5, 6**].

Thus a study was undertaken to investigate the effect of chitosan on the solubility of fenofibrate incorporated into this polymer carrier. Studies were carried out for different viscosity and average molecular weights of chitosan.

Demonstration of the effect of chitosan about average molecular weight in various formulations or with various methods of preparation of the solid dispersions on the dissolution of fenofibrate may enable development of new preparations of this drug with increased solubility.

2. Materials and methods

2.1. Materials

The study was performed with the use of fenofibrate (Fenofibrate p.a. min. 99%, SIGMA, Italy) incorporated into natural, highly purified chitosan **sample S** with 92% deacetylation and viscosity average molecular weight M_η=1087 kDa, intrinsic viscosity η = 0.7437 $dm^3\ g^{-1}$, **sample A** 92% deacetylation and viscosity average molecular weight M_η = 839 kDa, intrinsic viscosity η =0.5843 $dm^3\ g^{-1}$, **sample B** 92% deacetylation and viscosity average molecular weight M_η = 407 kDa, intrinsic viscosity η = 0.2986 $dm^3\ g^{-1}$

(Chitosan Huasu p.a., Chitin, France), sodium lauryl sulfate p.a., PPH „Stanlab", Poland, Aqua purification, acc. to FP VIII.

2.2. Methods

2.2.1. Examination of pure fenofibrate and its physical mixtures dissolution rate

Evaluation of dissolution was performed in a dissolution apparatus according to FPVII, which describes investigation of active substance solubility rate from solid drug forms [**7**]. The examination was performed in a VanKel VK 7025 dissolution apparatus, to which Varian Inc. fraction collector was attached. 1000 ml of 0.5% solution of sodium lauryl sulfate (SLS) at pH 6.8 was used as a release medium.

Solubility was evaluated after compressing 100 mg of samples, which were placed in each of the six chambers of the apparatus at 37 ± 0.5 °C, with velocity of 100 rotations per minute. The trial was continued for 1 hour, 5 ml samples were collected in 10 time intervals, i.e. after 5, 10, 15, 20, 25, 30, 35, 40, 50 and 60 minutes. Collected samples were filtered on filters with 10μm pore size.

The collected samples were diluted and next their content was evaluated with the use of JASCO V650 spectrophotometer with the use of 1 cm cuvette at wavelength $\lambda = 290$ nm.

The drug concentration in samples and an average percentage of dissolved fenofibrate were calculated using linear regression equation for fenofibrate, $y = 4389x + 0.0168$. Quantitative drug-to-polymer ratios in which the solid dispersion had the most beneficial properties improving the drug solubility were determined.

2.2.2. Technology for the preparation of investigated formulations

2.2.2.1. Preparation of samples for investigation of fenofibrate dissolution

The dissolution of fenofibrate was investigated immediately after compressing the powders in a Specac hydraulic press. The prepared 100 mg samples weighed on a Mettler balance were pressed on a punch die with a diameter of 13 mm. The pure drug and physical mixtures were pressed at a pressure of 5 ton for 20 sec.

2.2.2.2. Preparation of fenofibrate and polymer physical mixtures.

Physical mixtures were prepared by grinding in an agate mortar for 10 minutes of adequate amounts of fenofibrate with chitosan about average molecular weight: $M_\eta = 1087$ kDa, $M_\eta = 839$ kDa, $M_\eta = 407$ kDa at drug-to-polymer weight ratios 1:9, 3:7, 5:5 weighed on a Sartorius analytical balance. The prepared physical mixtures were passed through a sieve with 315 μm wholes, and next placed in glass bottles sealed with cork and stored in an exicator over silica gel. Every sample was prepared in 1 g amount (***Table 1***).

2.2.3. Statistical analysis

Statistical analysis was performed for maximum percentage values of fenofibrate solubility (in 60 minutes) from dispersions containing chitosan. The effects of chitosan as well as the effect of the dispersion preparation method on the drug solubility were analyzed. In order to check the normality of variables distribution, the following tests were performed: the Kolmogorov-Smirnov, the Lilliefors, as well as the Shapiro-Wilk tests. Next the variance homogeneity was

Table 1. *The quantitative composition of physical mixtures of the fenofibrate with chitosan.*

Average molecular weight of chitosan	Physical mixtures	Drug/ polimer ratio	Quantity of polimer, mg	Quantity of polimer, mg
Sample S M_η=1087 kDa	MFS55	5:5	500	500
	MFS37	3:7	300	700
	MFS19	1:9	100	900
Sample A M_η=839 kDa,	MFA55	5:5	500	500
	MFA37	3:7	300	700
	MFA19	1:9	100	900
Sample B M_η=407 kDa	MFB55	5:5	500	500
	MFB37	3:7	300	700
	MFB19	1:9	100	900

checked by means of the Brown-Forsythe's test at significance level $p < 0.50$ as well as ANOVA variance analysis was performed.

3. Results and discussion

Analysis of data included in ***Table 2 - 4*** and plotted dissolution curves presented in ***Figure 1*** revealed that the addition of chitosan had a beneficial effect on the fenofibrate dissolution profile in the investigated physical mixtures.

The results of the study demonstrated that all the investigated physical dispersions of fenofibrate with chitosan increased the solubility of fenofibrate.

The solubility findings of pure fenofibrate in 0.5% SLS were used as reference to compare the solubility of the drug incorporated into chitosan. The drug dissolution was found to increase gradually and it was from 0.49% in the time 5 min to 1.30% for 60 min of the investigated dose. The presence of chitosan increased significantly the solubility of fenofibrate, which increased with duration of the trial and with increasing percentage of polymer in the formulations.

The highest dissolution of fenofibrate, amounting to 86 %, was observed after 60 minutes from physical mixtures containing the drug-to-polymer weight ratio 1:9 in the presence of chitosan S. In dispersions containing 30% of the drug and of the polymer, the solubility of fenofibrate was at the level of 56, 66 %. The lowest solubility was observed in dispersions in which the drug-to-polymer weight ratio was 5:5, in which case the drug solubility was slightly above 15, 86%.

Comparing data from ***Tables 2*** and ***Figure 1***, we can notice a significant increase in the drug solubility, which in the presence of chitosan increased 13 times, 44 times and almost 70 times in relation to the amount of added polymer in comparison to the solubility of pure drug.

Table 2. *Influence of chitosan S (average molecular weight M_η = 1087 kDa) on the dissolution of fenofibrate from physical mixture in 0.5% aqueous solution SLS.*

Time, min	Fenofibrate - to - Chitosan S weight ratio of physical mixtures					
	MFS 5:5		MFS 3:7		MFS 1:9	
	average % of dissolved fenofibrate	standard deviation	average % of dissolved fenofibrate	standard deviation	average % of dissolved fenofibrate	standard deviation
5	0.76	0.10	7.33	0.73	23.35	0.07
10	2.65	0.27	12.87	1.30	47.52	1.15
15	3.88	0.49	21.41	2.80	60.63	2.16
20	5.26	0.65	26.14	2.45	70.00	2.74
25	6.92	0.62	30.06	0.18	76.13	3.45
30	8.30	0.71	35.21	0.42	79.77	3.76
35	10.44	0.17	38.95	0.54	83.77	2.63
40	12.17	0.54	43.05	0.65	85.57	4.01
50	13.61	1.04	50.11	0.86	85.09	1.54
60	15.86	1.03	56.66	0.13	86.00	1.29

Table 3. *Influence of chitosan A (average molecular weight M_η = 839 kDa) on the dissolution of fenofibrate from physical mixture in 0, 5% aqueous solution SLS.*

Time, min	Fenofibrate - to - Chitosan A weight ratio of physical mixtures					
	MFA 5:5		MFA 3:7		MFA 1:9	
	average % of dissolved fenofibrate	standard deviation	average % of dissolved fenofibrate	standard deviation	average % of dissolved fenofibrate	standard deviation
5	0.64	0.05	7.97	0.08	20.83	0.01
10	2.29	1.11	16.92	0.02	36.15	0.01
15	4.15	2.05	24.08	0.12	46.25	0.11
20	6.13	2.60	29.81	0.19	53.94	0.47
25	8.12	3.27	35.04	0.33	58.79	0.66
30	10.24	3.56	39.51	0.33	63.21	0.62
35	12.14	2.50	43.66	0.55	67.87	0.70
40	14.12	3.80	47.37	0.60	70.22	0.81
50	17.78	1.48	53.63	0.80	74.39	0.66
60	21.41	1.23	58.85	1.01	77.66	0.59

When the course of fenofibrate solubility curves in the presence of chitosan is observed, it becomes apparent that they are situated in the field above the fenofibrate solubility curve without polymer. The solubility curve for fenofibrate mixed with chitosan S at 1:9 ratio assumes the highest position in the field, also the inclination angle of the straight line to time axis is significant, and the drug solubility in relation to time is from 23.35% to 86.00%, i.e. it increases with time.

The solubility line of pure fenofibrate is characterized by a low inclination angle to the time axis, and the drug solubility in time increases slightly and is from 0.49% to 1.3%. Increased solubility of fenofibrate in physical mixtures with chitosan may be explained by numerous factors. Decreased size of the molecules may be the first of them. Chitosan, when

Table 4. *Influence of chitosan B (average molecular weight M_η = 407 kDa) on the dissolution of fenofibrate from physical mixture in 0.5% aqueous solution SLS.*

Time, min	Fenofibrate – to - Chitosan B weight ratio of physical mixtures					
	MFB 5:5		MFB 3:7		MFB 1:9	
	average % of dissolved fenofibrate	standard deviation	average % of dissolved fenofibrate	standard deviation	average % of dissolved fenofibrate	standard deviation
5	1.05	0.31	6.65	0.01	14.81	0.67
10	1.96	0.51	13.04	0.20	24.55	0.45
15	3.12	0.68	19.49	0.31	32.70	0.04
20	4.32	0.88	24.59	0.47	39.64	0.01
25	5.48	1.04	28.32	0.07	44.50	0.23
30	6.60	1.13	32.95	0.46	48.78	0.04
35	7.63	1.14	36.27	0.30	52.93	0.19
40	8.72	1.21	39.10	0.12	55.97	0.41
50	10.73	1.25	44.16	0.43	61.04	0.22
60	12.73	1.30	48.85	0.49	65.18	0.27

dispersing in water, may cause molecular dispersion of the drug by increasing the surface of the drug solubility [**8**].

Comparison of data from ***Tables 2 - 4*** and ***Figure 1*** demonstrates a significant increase in the drug solubility, which in the presence of chitosan - M_η = 1087 kDa (sample MFS) in dispersions was almost 58 times higher in relation to the amount of added polymer in comparison to the solubility of pure drug. It was noticed that the dissolution rate of fenofibrate from the dispersions 1:9 in the presence of chitosan - Mη = 839 kDa (sample MFA) and Mη = 407 kDa (sample MFB) was obviously lower than that from the dispersions with chitosan - Mη = 1087 kDa.

The enhancement of drug dissolution rate is 52-to 44-fold for weight ratio 1:9 solid dispersion fenofibrate in chitosan- Mη = 839 kDa and chitosan Mη = 407 kDa respectively formulations.

In dispersions containing 10% of the drug and 90% of the chitosan about average molecular weight 839 kDa , the solubility of fenofibrate was at the level of 77.6%. The lowest solubility was observed in dispersions in which the drug was incorporated in chitosan about average molecular weight 407 kDa in which case the drug solubility was slightly above 65%. The result demonstrated higher drug dissolution of the physical mixtures containing chitosan sample A than that of chitosan sample B and sample C.

Chitosan in dispersions may prevent agglomeration of fenofibrate molecules and increase wettability of the drug molecules, thus intensifying the drug solubility. The above findings were supported by statistical analysis of the investigated samples.

Normality tests performed for the percentage amount of dissolved fenofibrate after 60 minutes in relation to the drug-to-polymer weight ratio revealed normal distribution of data. Variance analysis demonstrated statistically significant differences in the findings in

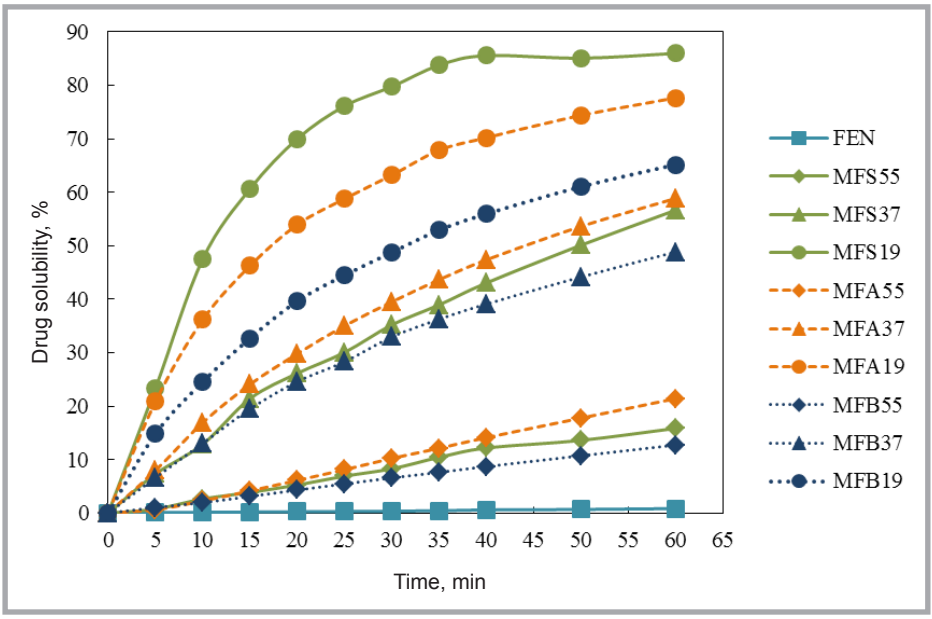

Figure 1. *Dissolution profiles of fenofibrate from physical mixture in aqueous solution 0.5% SLS medium.*

relation to the applied composition of physical mixtures. Value $p < 0.05$ was assumed as statistically significant.

4. Conclusions

1. Chitosan significantly increased the dissolution rate of fenofibrate.
2. The effect depends on the drug-to-polymer quantitative ratio. The highest solubility of fenofibrate was achieved in the presence of chitosan at the drug-to-polymer weight ratio 1:9 (10% of the drug and 90% of the polymer). The disolution enhancement in physical mixture of the drug-to-polymer weight ratio 1:9 was ~70 times higher than that of fenofibrate alone.
3. The effect depends on the preparation techniques of physical mixtures and on the molecular weight of chitosan. The dissolution rates of fenofibrate in the presence of chitosan at the weight ratio 1:9 increased with the increment of the molecular weight of the chitosan.
4. The above results may enable development of new technologies for fenofibrate formulations with the use of chitosan, which would be characterized by better solubility and thus increased bioavailability of the drug.

5. References

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