FABAD J. Pharm. Sci., 19, 97-102, 1994

Bioavailability of Phenytoin Products Marketed in Türkiye

Gül AYANOĞLU-DÜLGER*, Levent KABASAKAL*, Jülide AKBUĞA**, Recep ÖZVEREN***

Türkiye'deki Fenitoin Müstahzarlarından Biyoyararlanım

Özet: *Özellikle epilepsilerin ve aritmilerin tedavisinde kullanılan fenitoinin biyoyararlanım problemleri arzettiği bilinmektedir. Bu çalışmamızda Türkiye'de pazarlanmakta olan üç fenitoin müstahzarından yararlanımı karşılaştırmalı olarak ve rastgele çaprazlama yöntemine göre inceledik.*

Bunlardan iki tanesi tablet, diğeri ise kapsül şeklinde olup hepsi 100 mg dozunda fenitoin sodyum içermektedirler. Çalışma 8 sağlıklı gönüllüde yapılmış ve ilacın deneklere sabah aç karnına verilmesini takiben venöz kan örnekleri belli aralıklarla 24 saat süresince toplanmıştır. Plazma fenitoin konsantrasyonları No 9518-05 Abbott TDx Sistemi aracılığı ile ölçülmüştür. Her üç müstahzardan da biyoyararlanım bireyler arasında çok fazla değişkenlik göstermiş, ancak müstahzarların ortalama biyoyararlanımları arasında anlamlı bir fark bulunamamıştır. Müstahzarların içerdiği fenitoin sodyum miktarı spektrofotometrik olarak tayin edilmiş ve kabul edilebilir sınırlar içinde olduğu saptanmıştır. Müstahzarların in vitro dissolüsyon hızları USP XXII yöntemi ile incelendiğinde ise A (tablet) ve C (kapsül)'nin USP standartlarına uygun olduğu, B (tablet)'nin ise uymadığı ve yavaş çözündüğü, ayrıca agregatlar halinde sepette biriktiği gözlenmiştir.

Sonuç olarak, incelenen müstahzarların in vitro dissolüsyon özellikleri ile in vivo biyoyararlanımları arasında bir korelasyon bulunmadığı; ayrıca fenitoin tedavi indeksi dar ve doza bağımlı kinetik gösteren, kararlı durum plazma ilaç konsantrasyonları absorpsiyon nispetindeki değişmelerden fazlası ile etkilenen bir ilaç olduğu için, biyoyararlanımları çok fazla değişkenlik gösteren bu müstahzarların sürekli kullanılmaları sırasında istenmeyen bir durumun ortaya çıkmasını önlemek üzere hastanın yakından izlenmesinin gerektiği söylenebilir.

Anahtar sözcükler *: Fenitoin, Biyoyararlanım, Dissolüsyon*

Summary: *It is known that phenytoin is liable to bioavailability problems. We investigated the comparative bioavailabilities of three commercial products marketed in Turkey in a random, crossover study. Two of these were tablet forms and the third a capsul formula; all of them contained 100 mg of the drug as the sodium salt. The study was carried out in 8 healthy volunteers to whom the drug was administered after an overnight fast and venous blood samples were obtained for 24 hours. The amount of phenytoin in plasma was determined by the Abbott TDx system No 9518-05.*

There was no significant difference between the bioavailabilities of these products; ie they were bioequivalent; yet, both the rate and extent of absorption showed large interindividual variability for all three products. The amount of phenytoin in the products was determined spectrophotometrically and all of them were found to be within the acceptable limits. Thus, the variability in the total amount absorbed(AUCs) was not due to an inconsistency of the amount of active ingredient in the formulas. In vitro dissolution rates of the products were determined by the USP XXII method and two of them were found to comply with the given standards, but one of tha tablets(B). did not.

It was concluded that there was no correlation between the in vitro dissolution and in vivo absorption rates of these drugs, and that since phenytoin is a drug with a low therapeutic index and which shows nonlinear kinetics, patients who are being treated with any one of these products should be monitored closely in order to prevent any undesired outcome due to the large variability in their bioavailabilities.

Keywords *: Phenytoin, Bioavailabilit Dissolution*

Geliş tarihi *: 22.10.1992*

Kabul tarihi *: 17.5.1994*

(*) Marmara University, Sch. of Pharmacy, Dept. of Pharmacology, Haydarpaşa, İstanbul.
(**) Marmara University, Sch. of Pharmacy, Dept. Pharmaceutical Technology, Haydarpaşa, İstanbul.
(***) Kasımpaşa Marine Hospital, Dept. Pharmacology, Kasımpaşa, İstanbul.

Introduction

Phenytoin is widely used in the treatment of many types of convulsive disorders and some arrhythmias. It is a weak acid with a pKa value of 8.3 and is practically insoluble in water.

With these physicochemical characteristics it is to be expected that some difficulties will be encountered in dissolution of phenytoin in the gastrointestinal tract and therefore in its absorption. Thus, it has been classified as a drug with "high risk potential" with respect to bioavailability problems[1-3]. Many reports concerning the bioavailibility from various products of phenytoin have been published, but to our knowledge, the bioavailability of the products marketed in Türkiye has not been investigated before[1,2,4-6]. In a study by Arı-Ulubelen et al.[7], dissolution rates of phenytoin sodium tablets and capsules were determined in different lots of two commercial preparations and it was observed that both tablets and capsules show different dissolution profiles for different lots.

In this study, we evaluated the relative bioavailability of two tablet formulations and one capsule formula in a single dose, random cross-over study in healthy volunteers. We also determined the amount of active drug in the formulas and their dissolution rates.

Materials and Methods

The study was carried out on 9 healthy males (one had to drop out on the last day of the study, so that during the statistical evaluation the number of volunteers was regarded as 8); 19 to 21 years of age; weighing between 66 to 80 kg and who were informed about the nature of the study. Subjects received no other drugs or alcohol during the study, nor for at least a week preceding each study day. Volunteers were randomly assigned to 3 groups of 3 subjects each, each group receiving the same product on the same day. The experiment was performed three times, with each product in each subject, with a washout period of 4 weeks between studies. On each study day, a heparinized catheter was inserted into a superficial antecubital vein of each volunteer for blood sampling and the volunteers were kept at rest in the clinic for at least 12 hours.

In the morning, each volunteer received 100 mg of the drug after an overnight fast with 200 ml of water and was not allowed to eat or drink anything for 3 hours thereafter. Volunteers were given a standart meal 4 hours after the drug and then after the 8th hour blood sample was taken. Venous blood specimens were obtained immediately before dosing and at regular intervals for 24 hours after drug administration. Serum was separated and stored at -18°C until assayed.

During the first study period, blood samples were collected for 48 hours, but since no drug was found in any of the samples after 24 hours, blood sampling was terminated at 24 hours thereafter.

Serum phenytoin levels were determined in duplicates by the Abbott TDx system No 9518-05. Abbott TDx system is a fluorescent polarisation immunoassay(FPIA), in which special kits are used to measure drug concentration in serum. Results obtained with this method were again controlled with the spectrophotometric method. Relative bioavailability was determined by using the AUC_{0-24} (area under the serum concentration-time curve), peak serum concentration (C_{max}) and the time to reach C_{max} (T_{max}) as the basic parameters. AUC was calculated using the trapezoid rule. Statistical method used was the two way analysis of variance.

For the dissolution studies, the rotating basket method of the USP XXII was used. The studies were carried out at 37±0.10°C in 900 mL deaerated distilled water with the basket rotating at 50 rpm (ElektroMag). The samples were taken at regular time intervals and the drug content was spectrophotometrically determined at 234 nm(Varian UV-2100). At least 6 tablets were used for each determination. The amount of phenytoin in the products was also determined, using 10 tablets or capsules for each product. In both cases, the same spectrophotometric method of Olesen[8] was used to determine the amount of phenytoin.

Results

Mean phenytoin serum concentration versus time curves, following oral administration of 100 mg each of 3 different products of phenytoin sodium

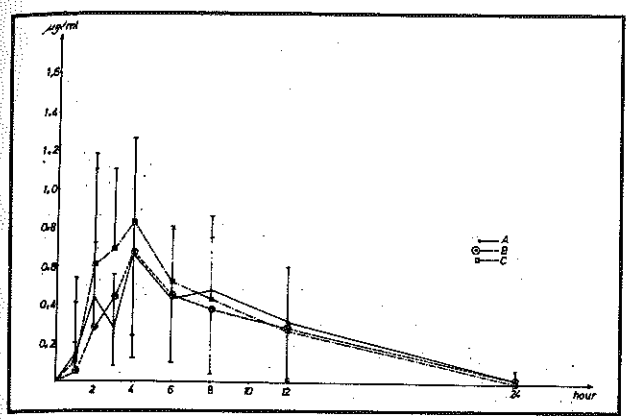

Figure 1. The mean phenytoin plasma concentration versus time curves for the three products investigated.

are presented in Fig 1. Though it seemed as if product C had a better bioavailability than the tablet formulations, there was no statistically significant difference between the bioavailability of 3 products expressed in terms of either AUC_{0-24} or rate of absorption (Table 3).

	Products		
	A	B	C
Registration Number	113/16	143/67	141/23
Registration Date	03.11.1972	14.12.1987	11.02.1987
Serial No.	52	101504	009087
Production Date	11.1990	1990	09.1990
Form	Tablet	Tablet	Capsule

Table 1. Total bioavailability (AUC 0-24 µg.hr/mL) of each product.

	Product		
Volunteer	A	B	C
C.K.	2.65	0.42	11.73
N.Ö.	3.38	1.88	1.4
M.I.	1.60	12.25	7.17
M.Ö.	5.15	1.74	9.45
B.D.	6.81	7.7	5.28
İ.P.	14.59	7.85	13.54
M.A.Ö.	14.36	3.61	7.27
A.D.	4.83	14.01	1.27
Mean	6.67	6.18	7.14
S.D.	5.07	5.09	4.44
S.E.	1.79	1.8	1.57
Confidence Int. (% 95)	2.44<u<10.90	1.93<u<10.43	3.44<u<10.85

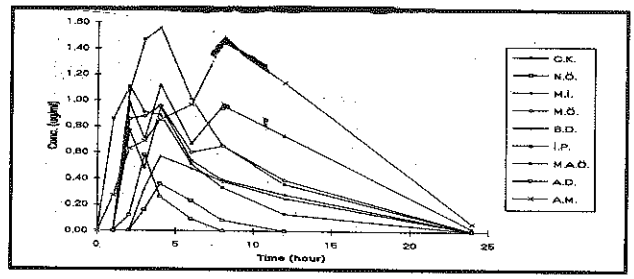

Figure 2. Plasma concentration versus time curves obtained following administration of a single oral dose of product C, the capsule formula, to 9 volunteers.

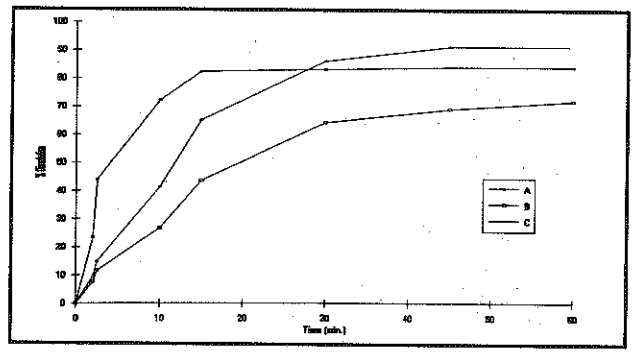

Figure 3. Dissolution profiles of the three phenytoin products.

Both the rate and the extent of absorption of all the products showed large interindividual variability(Table 1 and 2). Fig. 2, which shows the serum levels of phenytoin after administration of product C, (which seemed to be the best among the three) to the volunteers is an example for this variability.

The amount of phenytoin sodium in each formulation was determined and was found to be within acceptable limits(Table 4).

Dissolution profiles of commercial phenytoin preparations are shown in Fig 3 and the results are summarized in Table 5 and 6. Data were evaluated kinetically. Dissolution process followed first order kinetics. Although they contained analytically the same amount of the drug, brand A showed the highest dissolution rate, while B was the slowest. In B, tablet contents did not dissolve completely in the dissolution medium in one hour and remained in aggregates. The rank order for the dissolution rates of these products was A>C>B. Brands A and C met the USP requirements for the dissolution, but B did not.

Table 2. T_{max} (hr) and C_{max} (μg/mL) values of each product.

	A		B		C	
Volunteer	T_{max}	C_{max}	T_{max}	C_{max}	T_{max}	C_{max}
C.K.	4	0.28	4	0.28	4	1.56
N.Ö.	4	0.78	2	0.45	4	0.36
M.I.	12	0.20	4	1.15	4	0.95
M.Ö.	2	0.59	4	0.38	4	096
B.D.	6	0.76	3	0.93	4	0.57
İ.P.	8	1.19	4	0.69	4	1.12
M.A.Ö.	2	2.15	6	0.40	2	1.11
A.D.	4	0.82	4	1.45	3	0.58
Mean	5	0.85	4	0.72	4	0.9
S.D.	3	0.61	1	0.42	1	0.38
S.E.	1	0.22	0	0.15	0	0.13
Confidence Int. (% 95)	3<u<8	0.33<u<1.4	3<u<5	0.37<u<1.08	3<u<4	0.58<u<1.22

Table 3. Analysis of Variance: Comparison of A, B and C.

Variable	Sum of Squares	Degrees of Freedom	Mean Square	F	P
Between Volunteers	167.44	7	23.92	1.44	p>0.25
Between Periods	103.22	2	51.61	3.1	0.10>p>0.05
Error	232.76	14	16.63		
Total	503.41	23			
Between Volunteers	167.37	7	23.91	1.01	p>0.25
Between Products	3.68	2	1.84	0.08	p>0.25
Error	332.57	14	23.76		
Total	503.61	23			

Discussion

Phenytoin is a weak acid, which is poorly soluble in water. Although phenytoin sodium is readily soluble in water, in the acidic medium of the stomach it also precipitates subsequent to its dissolution from a solid dosage form. Thus, it has been claimed that the biovailability of phenytoin sodium and phenytoic acid are the same and that the size of acid crystals, aggregates or particles entering the intestine is probably the most critical factor in determining the rate and extent of absorption[1-3]. Thus, whether the acid or the sodium salt is administered, the absorption of the drug will depend upon the product formulation[3]. Besides, although phenytoin is mainly absorbed from the proximal part of the small intestine, it has also been reported that the rate of absorption may be rather slow and variable[1]. Thus, the wide interindividual variation of the rate and extent of absorption of phenytoin products may be attributable to its own physicochemical properties as well as the formulation factors.

In a previous study, in which the in vitro dissolution characteristics of A and C were evaluated and dissolution rates were found to be different for different lots of the products and the slow dissolution rates were attributed to aggregates observed in the basket[7]. However, in this study, brand A and C met the USP XII requirements for dissolution(for the lots used in the study). Brand B had the slowest rate of dissolution and the tablet contents remained in the medium as aggregates. The slow and variable absorption profile of brand B could have been attributed to the slow and incomplete dissolution, had the other products shown better absorption profiles; but the bioavailability of brands A

Table 4. The Amount of Phenytoin Sodium in the Commercial Products.

	Product (mg/tab)		
	A	B	C
	100	102	100
	100	102	88.8
	100	102	102
	100	100	102
	102	100	102
	100	100	98.6
	100	103	103
	103	100	100
	100	103	103
	103	100	100
Mean	101	101	99.9
S.D.	1	1	4.2

Table 5. Determination Coefficents (r^2) of Products.

	Product		
Kinetic Model	A	B	C
Zero order	0.667	0.952	0.919
First order	0.871	0.966	0.984
R.R.S.B.W.	0.662	0.714	0.749
Hixson-Crowell	0.690	0.438	0.398

Table 6. Kinetic parameters for the first order dissolution of Phenytoin Products.

Product	t (min)	k (min-1)	A (%)
A	9.9	0.0700	83.5
B	24.3	0.0285	65.2
C	11.6	0.0597	86.3

* Percent dissolved in 30 min.

and C were not significantly different from that of brand B.

When in vivo absorption profiles of these products were studied, although the mean C_{max} and the mean AUC_{0-24} values for Brand C(the capsule formula) seemed larger, no statistically significant difference between these parameters for the products A, B and C was found. Thus, it may be claimed that these three phenytoin products marketed in Türkiye are bioequivalent. On the other hand, the large interindividiual variability observed in the bioavailability of each of these products raises the possibility that there will also be large intraindividual variance during treatment of epileptics with any of these products.

In general, it is known that different brands of a certain drug may have differences in their bioavailability. Because of this, it is recommended that especially during treatment with drugs with a low therapeutic index or those that are used for life threatening conditions (such as heart failure or epilepsia), changes in dosage form or manufacturer should be avoided once a patient's dosage requirements are established, as a relatively small decrease or increase in bioavailability can greatly alter the plateau plasma concentration during chronic administration[3,4]. These are also true for phenytoin. Besides, phenytoin shows capacity-limited metabolism(dose-dependent elimination kinetics) and this can be observed in many patients already in the therapeutic concentration range; therefore even small changes in drug dosage(or bioavailability) could be expected to considerably alter serum steady state phenytoin concentrations[1,3,4,6,9]. Because of this, existence of phenytoin products with different bioavailability is a serious clinical problem. The importance of continuing treatment with one brand and dosage form of phenytoin, once adequate seizure control has been obtained, has often been stated, but with the presently observed inter-individual variability in bioavalability of the same brand, it would be difficult to attain a steady state even with regular dosing; because, theoretically, although significant alterations in absorption rates should not affect average steady state levels, the alterations in the extent of absorption will[9-11]. In this study, both the rate and extent of absorption of all the products varied significantly.

We did not observe a good correlation between the in vivo and vitro data, however, in a previous study it has also been reported that there is no correlation between in vivo and in vitro data[2].

It may be concluded that the in vitro dissolution characteristics of a formulation is not always a good indicator of in vivo absorption; thus, it should not be used as a guide to determine the bioavailability of a certain formulation. Since phenytoin is a drug with a low therapeutic index and which

shows dose-dependent kinetics, patients who are being treated with any of these products marketed in Türkiye should be monitored closely in order to prevent any undesirable outcome due to large variability in their bioavailability.

References

1. Neuvonen P. J., "Bioavailability of Phenytoin; Clinical Pharmacokinetics and Therapeutic Implications" *Clin. Pharmacokin.*, 4, 91-103, 1979.
2. Arnold, K., Gerber, N., Levy G., "Absorption and Dissolution Studies on Sodium Diphenylhydantoin Capsules", Canad. *J. Pharmaceut. Sci.*, 5, 89-92, 1970.
3. Tozer T. N., Winter M. E., "Phenytoin", in Evans W. E., Schentag J. J., Jusco W. J. (eds), 'Applied Pharmacokinetics', San Francisco, *Applied Therapeutics Inc.*, pp. 275, 1981.
4. Gibaldi M., "Bioavailability", In: *Biopharmaceutics and Clinical Pharmacokinetics*, 4th Ed., Philadelphia, London, Lea and Febiger, pp. 168, 1991.
5. Matsukura, M., Ikeda, T., Higashi, A., Matsuda, I., "Relative Bioavailability of Two Different Phenytoin Preparations', *Dev. Pharmacol.*, 7, 160-168, 1984.
6. Sawchuk, R. C., Pepin, S. M., Leppik, I. E., Gumnit, R. J., "Rapid and Slow Release Phenytoin in Epileptic Patients at Steady State: Comparative Plasma Levels and Toxicity", *J. Pharmacokin. Biopharm.* 10, 365-382, 1982.
7. Arı-Ulubelen, A. Akbuğa J., Bayraktar-Alpmen G., "Dissolution Studies on Phenytoin Sodium Tablets and Capsules", *Sci. Pharm.* 51, 191-197, 1983.
8. Olesen. O. V., "Determination of Phenobarbital and Phenytoin in Serum by Ultraviolet Spectrophotometry", *Scand. J. Clin. Lab. Invest.*, 20, 63-69, 1967.
9. Sawchuk, R. J., Rector T. S., "Steady-State Plasma Concentrations as a Function of the Absorption Rate and Dosing Interval for Drugs Exhibiting Concentration-Dependent Clearance: Consequences for Phenytoin Therapy", *J. Pharmacokin. Biopharm.* 7, 543-555, 1979.
10. Neuvonen, P. J., Bardy A. ve Lehtovaara R., "Effect of Increased Bioavailability of Phenytoin Tablets on Serum Phenytoin Concentration in Epileptic Out-Patients" *Br. J. Clin. Pharmacol.* 8, 37-41, 1979.
11. Sawchuk, R. J., Pepin, S. M., Leppik, I. E., Gumnit, R. J., "Rapid and Slow Release Phenytoin in Epileptic Patients at Steady State: Assesment of Relative Bioavailability Utilizing Michaelis-Menten Parameters", *J. Pharmacokin. Biopharm.*, 10, 383-391, 1982.