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Assessment of a hydroalcoholic surfactant solution as a medium for the dissolution testing of phenytoin

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Abstract

A buffered hydroalcoholic surfactant solution has been developed as a dissolution medium for phenytoin based on a statistically designed solubility study. A compromise between acceptable composition (relatively low additive concentrations and physiologically relevant pH), sufficient phenytoin solubility at a pH lower than the drug's pKa (8.3) and potential discrimination power was considered in the design. The medium selected was a pH 6.8 phosphate buffer containing 12% alcohol and 0.4% Tween 80. Both phenytoin and phenytoin sodium bulk powder dissolved at a higher rate in the test medium relative to water (the USP medium for phenytoin sodium capsules). The medium proved sensitive to dissolution differences between phenytoin sodium samples with different content of the acid form. A linear relationship was obtained between the acid content and percent dissolution at 30 min for these samples. Further, the test medium could detect the reduction in dissolution of a phenytoin sodium sample subjected to storage conditions (1 month at 40°C and 75% R.H.) allowing partial conversion to the acid form. From a practical standpoint, these findings may be useful in assessing the in vitro performance of both phenytoin and phenytoin sodium formulations and their dissolution stability.

Keywords: Phenytoin; Dissolution; Central composite design; Hydroalcoholic surfactant solutions

1. Introduction

Phenytoin (diphenylhydantoin) is a widely used anticonvulsant for the treatment of grand mal epileptic seizures. It is administered either as the acid form or the sodium salt. Significant variations in the in vitro availability of phenytoin as well as phenytoin sodium from commercial oral solid dosage forms have been reported and attributed to pharmaceutical and formulation factors (Bastami and Groves, 1978; Shah et al., 1983; Ari-Ulubelen et al., 1986; Yakou et al., 1986). Further, a considerable reduction in drug release was observed upon storage of phenytoin sodium capsules (Rubino et al., 1985; Abdallah et al.,

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1989), probably due to absorption of moisture and carbon dioxide with subsequent conversion of phenytoin sodium to the insoluble acid form (Gupta and Gupta, 1979).

In view of such variations, dissolution criteria have been set up by the USP for phenytoin sodium capsules using water as the dissolution medium (USP 23 – NF 18, 1995). However, there is no compendial dissolution requirements for phenytoin and no dissolution medium has been recommended for the comparison of the release characteristics of phenytoin and phenytoin sodium products. The use of water as a dissolution medium for these products resulted in a discrepancy between in vitro release and in vivo performance data (Brandau and Wehnert, 1979; Sved et al., 1979; Stavchansky and Gowan, 1984). Because of the practical insolubility of phenytoin and its fairly high pKa value, 8.3 (Philip et al., 1984), its dissolution in water or at physiological pH values is very slow (Serajuddin and Jarowski, 1993). Phenytoin sodium exhibits a high dissolution rate in water as a result of the self-buffering effect of the dissolved salt with consequent increase in pH and drug solubility (Serajuddin and Jarowski, 1993). However, under gastrointestinal pH conditions (pH 2–8), the sodium salt is rapidly converted to the practically insoluble acid form, an effect not reproduced in vitro when water is used as the dissolution medium. Thus, due to such a conversion either in the physiological milieu or at the surface or inside solid oral dosage forms upon storage, a dissolution medium in which phenytoin is sufficiently soluble may produce more discriminative data in the dissolution testing of phenytoin and phenytoin sodium.

Attempts have been made to increase the solubility of phenytoin in the dissolution medium by periodically replacing the medium during the dissolution run (Serajuddin and Jarowski, 1993) or using alkaline buffers allowing ionization of the drug (Neuvonen et al., 1977 and Brandau and Wehnert, 1979). Because of the relatively large dose of the drug and the alkalinity of the medium (pH > 8.3), these approaches seem either cumbersome or physiologically irrelevant.

In an earlier study (El-Massik et al., 1995), a solution buffered at pH 7.4 and containing a relatively low percentage of alcohol and a non-ionic surfactant (Tween 80) has been proposed for the dissolution testing of the water insoluble drug glibenclamide. This solution offered a compromise between practical and physiological considerations and exhibited a high discriminative power. In the present study, the applicability of the same approach to the design of a dissolution medium for phenytoin is assessed in terms of phenytoin solubilization and dissolution discrimination potential.

2. Materials and methods

2.1. Materials

Phenytoin and phenytoin sodium were kindly supplied by the Nile Company for Pharmaceuticals, Cairo, Egypt. Tween 80 (BDH Chemicals Ltd, Poole. England), methanol (HPLC grade, Romil Chemicals Ltd), ethanol and potassium dihydrogen orthophosphate (El-Nasr Pharmaceutical Chemicals Co., Egypt) were used in the study.

2.2. Solubility study

The equilibrium solubility of phenytoin was determined at 37°C as a function of pH of 0.1 M phosphate buffer, alcohol and Tween 80 concentrations as three independent variables applied simultaneously. A central composite optimization design (Bayne and Rubin, 1986) was used to select the level of the three variables in combinations required by the design. These are shown in Table 1. Excess amounts of phenytoin were added to 10 ml of the different media in stoppered conical flasks. The mixtures were shaken for 24 h and equilibrated for 12 h. in a constant temperature water bath. Samples filtered through a 0.22 μm Millipore filter were immediately diluted with the mobile phase, (methanol water, 6:4) and assayed by HPLC.

Table 1
Central composite design for the solubility study and observed response values (phenytoin solubility, mg/100 ml at 37°C)

Trial	pH	% Alcohol	% Tween 80	Solubility, mg/100 ml
1	6.8.	13.5	0.38	17.90
2	6.8	13.5	0.10	14.04
3	6.8	3.5	0.38	11.13
4	6.8	3.5	0.10	6.43
5	4.8	13.5	0.38	13.75
6	4.8	13.5	0.10	11.91
7	4.8	3.5	0.38	9.00
8	4.8	3.5	0.10	6.10
9	7.5	8.5	0.24	14.52
10	4.1	8.5	0.24	10.55
11	5.8	17.0	0.24	19.71
12	5.8	0.0	0.24	7.26
13	5.8	8.5	0.48	12.87
14	5.8	8.5	0.00	7.55
15	5.8	8.5	0.24	9.97
16	5.8	8.5	0.24	9.78
17	5.8	8.5	0.24	10.16

2.3. Phenytoin analysis

Phenytoin analysis was performed using essentially the USP-23 high pressure liquid chromatographic assay of phenytoin. The chromatographic system consisted of a constant flow pump (waters Model 501), a septumless injector (Waters Model U6K), a C18-Bondapack column (3.9 mm × 30 cm) that contains 10 μm packing L1, a tunable spectrophotometric detector (Waters Model 486) and a data module (Waters Model 746). The mobile phase was a methanol/water mixture 6:4 flowing at a rate of 1.2 ml/min. The retention time of phenytoin was 4 min. The drug was detected at 254 nm.

2.4. Dissolution rate study

Dissolution experiments were performed using USP apparatus II (Hanson Research, Northridge, CA, USA) at 37°C at a paddle speed of 50 rev./min. The dissolution medium was 900 ml of either water or the test medium, selected on the basis of the solubility data obtained (0.1 M phosphate buffer pH 6.8 containing 12% alcohol and 0.4% Tween 80). Samples of the dissolution medium were filtered through 0.22 μm Millipore filter and analyzed for phenytoin using HPLC.

This procedure was used to test the dissolution of bulk powder samples (100 mg, particle size < 200 μm) of phenytoin, fresh phenytoin sodium, physical mixtures of phenytoin and phenytoin sodium containing 26% and 45% of the acid as well as phenytoin sodium stored for 1 month at 40°C and 75% relative humidity (phenytoin acid content 45%). Results presented are the average of three experiments.

The approximate phenytoin content of phenytoin sodium samples used in the dissolution study was determined by replicate titration of dispersions of 0.5 g of the sample in 50 ml distilled water against 0.1 N NaOH. The end point was the disappearance of turbidity.

3. Results and discussion

It has been reported that a combination of co-solvents and solubilizers whether surfactant or non-surfactant may result in a synergistic solubilizing action for some water insoluble drugs (El-Khordagui, 1991; El-Massik et al., 1995). The phenomenon has been made use of in the development of a dissolution medium for glibenclamide (El-Massik et al., 1995). The medium proposed

was a phosphate buffer, pH 7.4, containing alcohol and a non-ionic surfactant (Tween 80) at relatively low levels, 8.5% and 0.24% respectively. In order to utilize this system for the dissolution testing of phenytoin, the levels of the system variables have to be optimized to provide a medium with sufficient solubilizing power for phenytoin (pKa = 8.3) at a physiologically relevant pH. Optimization of phenytoin solubility in the medium was achieved by using a central composite experimental design for three factors (Bayne and Rubin, 1986). The values of phenytoin solubility in the 17 trials selected are shown in Table 1. These were correlated to levels of pH, alcohol and Tween 80 concentrations using the general linear model procedure (SAS/Stat User's Guide, 1988). Data fitting was performed to generate the following complete second order polynomial equation:

$$Y = 308.0 - 90.0\,X_1 - 4.7\,X_2 - 34.8\,X_3 + 7.3\,X_1^2 + 0.35X_2^2 - 39.2X_3^2 + 1.0X_1X_2 + 34.1X_1X_3 - 3.39X_2X_3 \quad (1)$$

$$P < 0.05$$

$$r^2 = 0.9882$$

Where Y is the response level (phenytoin solubility), X_1, X_2 and X_3 are levels of pH, alcohol and Tween 80 concentrations respectively.

A simplified polynomial equation including the most significantly ($P < 0.05$) contributing factors and their interactions was also generated (reduced model) using a stepwise regression procedure (SAS/Stat User's Guide, 1988) as follows:

$$Y = 305.1 - 88.5\,X_1 - 5.7\,X_2 + 7.5\,X_1^2 + 0.4\,X_2^2 + 1.0\,X_1X_2 + 20.1\,X_1X_3 \quad (2)$$

$$P < 0.05$$

$$r^2 = 0.9845$$

The above equation indicates that pH had the highest impact on phenytoin solubility. Although the pH range studied (4.1–7.5) was below the pKa of phenytoin (8.3), increasing the medium pH resulted in increased solubility. This is consistent with previously reported pH-solubility data for phenytoin (Serajuddin and Jarowski, 1993), which indicated a pH-independent solubility up to pH 6 and a solubility increase at higher pH values.

Table 2
Observed and predicted values of phenytoin solubility, mg/100 ml at 37°C at selected factor combinations from Table 1

Trial	Observed response value	Predicted response value
1	17.90	18.41
2	14.04	14.49
7	9.00	9.26
10	10.55	10.14
14	7.55	7.10

Both the full and reduced models provided excellent prediction of phenytoin solubility at selected levels of the independent variables. Data obtained using the reduced model are presented in Table 2. The model-predicted solubilities were very close to the corresponding experimentally determined values, indicating validity of the model.

Further, response surface graphs were generated for phenytoin solubility as a function of the independent variables (Figs. 1–3). The drug solubility increased almost steadily as the pH, alcohol and Tween 80 concentrations increased. Partial ionization of phenytoin as the medium pH approaches the pKa value would increase the polarity of the drug molecules, possibly increasing

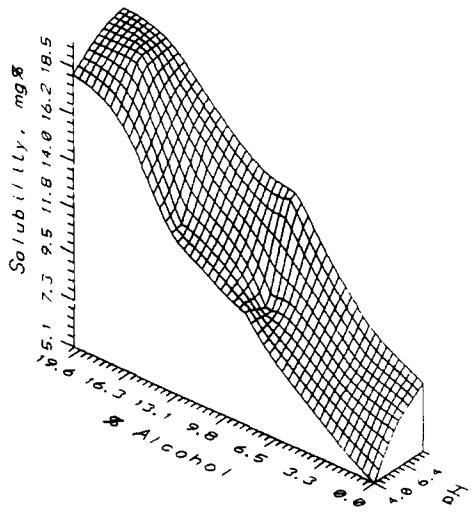

Fig. 1. Response surface graph relating phenytoin solubility to pH values and percent alcohol.

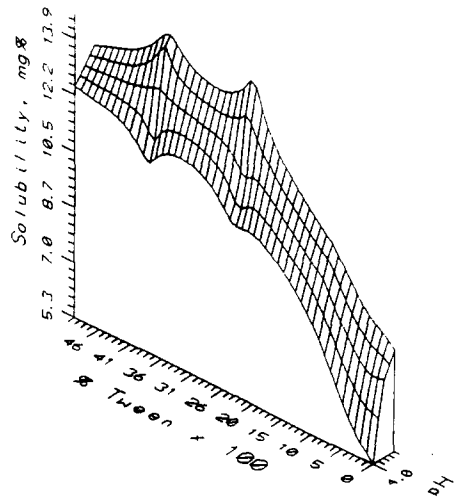

Fig. 2. Response surface graph relating phenytoin solubility to pH values and percent Tween 80.

solubility in alcohol and facilitating micellar incorporation. Such a view is supported by the positive interaction between pH and alcohol and pH and Tween 80, the latter being more pronounced, as indicated by the respective coefficients in Eq. 2. Change in the solubilizate polarity was shown to influence the amount solubilized and the location of solubilizate in the micellar structure (Mukerjee, 1971; Collett and Tobin, 1979).

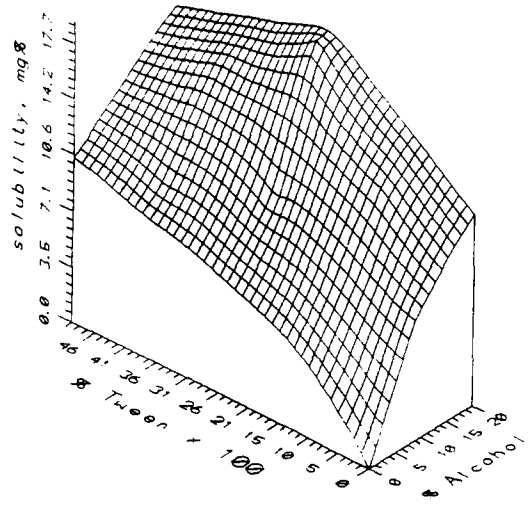

Fig. 3. Response surface graph relating phenytoin solubility to percent Tween 80 and percent alcohol.

Considering the objectives of this work, an optimum medium for the dissolution testing of phenytoin would be a system which provides sufficient solubility of the drug at a physiological pH (below the drugs pKa) and relatively low additive concentrations. Inclusion of alcohol at the high concentration levels (up to 40%) usually used in hydroalcoholic dissolution media, (Walkling et al., 1979 and USP 23-NF 18, 1995) was reported to impair tablet dissolution as a result of increase in disintegration time and suppression of the solubility of water soluble tablet excipients (Dodge and Gould, 1987 and Corrigan, 1991). Moreover, interference of alcohol with non-ionic micelle formation was shown to be limited at relatively low alcohol concentrations (Nishikido et al., 1974). Tween 80 was also kept at a relatively low post-micellar concentration to minimize the potential disintegration retarding effect reported for some formulations (Heng and Wan, 1985 and Pandit et al., 1985).

Based on the response surface graphs obtained, a system consisting of 12% alcohol and 0.4% Tween 80 in phosphate buffer pH 6.8 was selected as a medium for the dissolution testing of phenytoin. The drug solubility at 37°C in this medium was ~ 18 mg/100 ml compared to 3.5 mg/100 ml in water and ~ 4.0 mg/100 ml at pH 6 (Serajuddin and Jarowski, 1993).

Fig. 4 shows the dissolution profiles of phenytoin in the medium selected (test medium) and in water. An increase in both the rate and extent of phenytoin dissolution was observed in the test medium. While the profile in water levelled off at 60 min, dissolution of phenytoin continued in the test medium over the 2-h study period, probably as a result of uptake of dissolved phenytoin by the surfactant micelles. This might bear a similarity to the simultaneous drug release and absorption under physiological conditions, which appears to be an advantage offered by the medium proposed. However, the increase in dissolution rate is less than expected from the solubility data obtained. This phenomenon has been previously attributed to the lower diffusion coefficient of micelle-solubilised solutes in dissolution media containing surfactants (Itai et al., 1985; Parrot and Sharma, 1967)

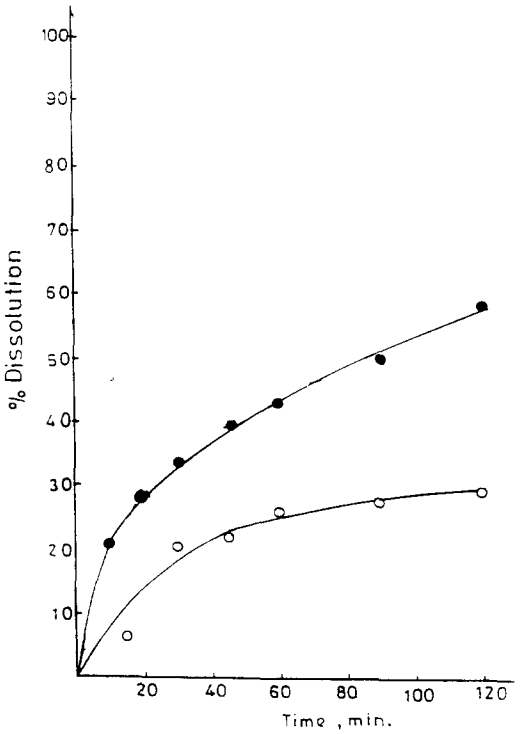

Fig. 4. Dissolution profiles of phenytoin acid (○) in water and (●) in phosphate buffer pH 6.8 containing 12% alcohol and 0.4% Tween 80 at 37°C.

The dissolution profiles of phenytoin sodium in the test medium compared to water are shown in Fig. 5. Complete dissolution was achieved in 30 min in the test medium and at about 2 h in water. Despite the increase in water pH as a result of the self-buffering action of phenytoin sodium (Serajuddin and Jarowski, 1993), the dissolution rate of the salt in water was lower than expected. The phenytoin sodium sample used was found to contain about 16% of the acid form This explains the dissolution behaviour of the salt observed in water.

Although the pH of the test medium (6.8) was not changed upon dissolution of phenytoin sodium, no precipitation of the free acid (pKa = 8.3) took place. Crystallization of phenytoin acid in the dissolution medium was reported previously under physiological pH-stat conditions (Serajuddin and Jarowski, 1993). This can be accounted for by the solubilization of unionized phenytoin in the test medium.

The discrimination power of the test medium was assessed by testing the dissolution of phenytoin sodium samples with different content of free acid (16%, 26% and 45%) in addition to a sample of phenytoin acid. A perfect rank order correlation (r = 0.9769) was obtained between the acid content and the % dissolution at 30 min (Fig. 6), indicating sensitivity of the test medium to differences in the dissolution properties of phenytoin samples which have undergone different degrees of conversion to the acid form. This may be useful in testing the in vitro performance of phenytoin sodium formulations in the production phase and upon storage. The discriminatory power of the test medium was further supported by the results of the dissolution

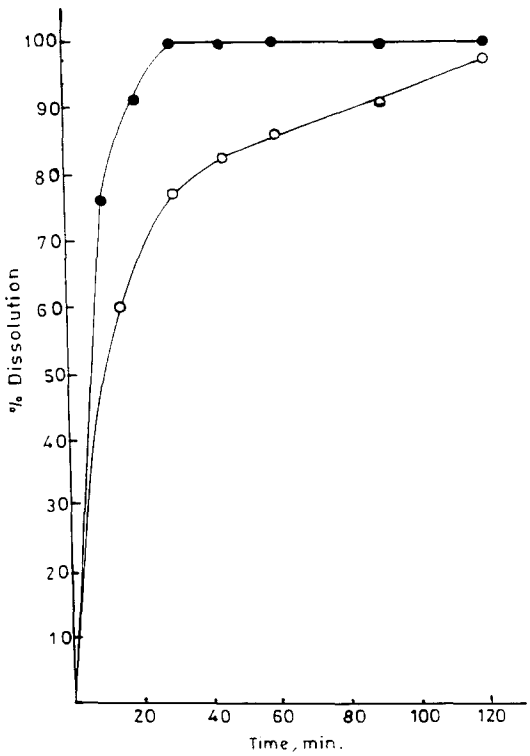

Fig. 5. Dissolution profiles of phenytoin sodium (○) in water and (●) in phosphate buffer pH 6.8 containing 12% alcohol and 0.4% Tween 80 at 37°C.

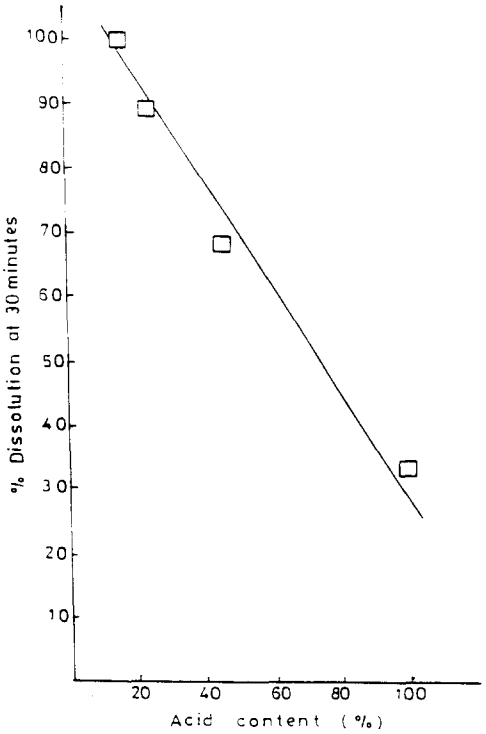

Fig. 6. Effect of phenytoin acid content on the percent dissolution of phenytoin sodium at 30 min in phosphate buffer pH 6.8 containing 12% alcohol and 0.4% Tween 80 at 37°C.

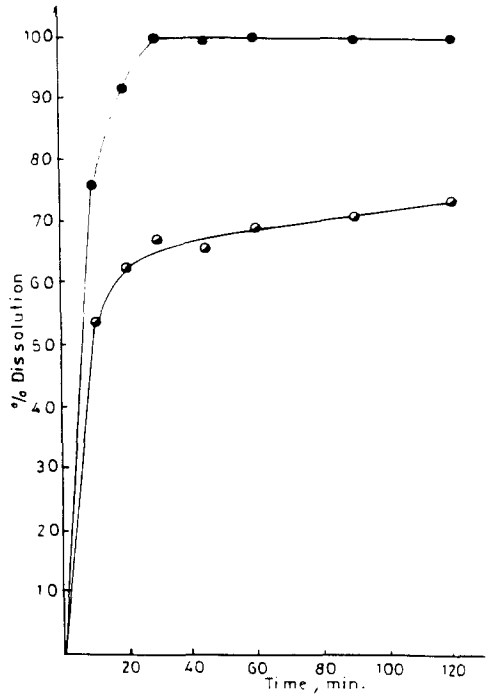

Fig. 7. Dissolution profiles of two phenytoin sodium samples in phosphate buffer pH 6.8 containing 12% alcohol and 0.4% Tween 80 at 37°C. (●) Fresh phenytoin sodium sample (acid content 16%). (◑) Phenytoin sodium stored for one month at 40°C and 75% relative humidity (acid content ~45%).

testing of a fresh sample of phenytoin sodium (acid content 16%) and a sample of the salt stored for one month at 40°C and 75% relative humidity (acid content 45%). A 32% reduction in the extent of dissolution of the stored sample was observed as a result of partial conversion to the acid form (Fig. 7).

In conclusion, the hydroalcoholic surfactant solution developed in this study offers promises as a dissolution medium for phenytoin and phenytoin sodium. The medium allowed dissolution of both chemical forms as well as phenytoin sodium samples with a different content of the acid form at a physiologically relevant pH with a good discrimination power, which are important prerequisites of a dissolution medium for phenytoin. These findings may be of value in assessing the uniformity and dissolution stability (Murthy and Ghebre-Sellasie, 1993) of phenytoin formulations. However, further studies are needed to correlate in vitro results obtained using this medium with bioavailability data.

References

Abdallah, O.Y., Ghaly, G.M., El-Khordagui, L.K., Naggar, V.F. and Khalil, S.A., A study of the in-vitro availability of phenytoin from solid dosage forms marketed in Egypt. *Alex. J. Pharm. Sci.*, 3 (1989) 151–155.

Ari-Ulubelen, A., Akbuga, J., Bayraktar-Alpmen, G. and Gülhan, S., Effect of formulation factors on the in-vitro dissolution characteristics of phenytion sodium capsules. *Pharm. Ind.*, 48 (1986) 393–395.

Bastami, S.M. and Groves, M.J., Some factors influencing the in-vitro release of phenytion from formulations. *Int. J. Pharm.*, 1 (1978) 151–164,

Bayne, C.K. and Rubin, I.B., *Practical experimental designs and optimization methods for chemists*, VCH Publishers, Beach Florida, 1986, pp. 150–152.

Brandau, R. and Wehnert, H.V., Dissolution rates and bioavailabiliy of phenytoin preparations. *Arzneim-Forsch. Drug Res.*, 29 (1979) 552–555.

Collett, J.H. and Tobin, E.A., Relationships between poloxamer structure and the solubilization of some para-substituted acetanilides. *J. Pharm. Pharmacol.*, 31 (1979) 174–177.

Corrigan, O.I., Co-solvent systems in dissolution testing: Theoretical considerations. *Drug Dev. Ind. Pharm.*, 17 (1991) 695–708.

Dodge, A. and Gould, P.I., Dissolution of chlorpropamide tablets in a methanol-water binary solvent system. *Drug Dev. Ind. Pharm.*, 13 (1987) 1817–1826.

El-Khordagui, L.K., A study of hydrotrope-cosolvent solubilised systems. *Alex. J. Pharm. Sci.*, 5 (1991) 103–108.

El-Massik, M.A., Darwish, I.A., Hassan, E.E. and El-Khordagui, L.K., Development of a dissolution medium for glibenclamide. *Int. Conf Pharm. Sci.* Tech., Alexandria, Egypt, March 22–25, 1995.

Gupta, V.Das and Gupta, A., Stability of some oral solid drug products when stored in counting machine. *Am. J. Hosp. Pharm.*, 36 (1979) 1539–1541.

Heng, W.S. and Wan, L.S.C., Surfactant effect on the dissolution of sulfanilamide granules. *J. Pharm. Sci.*, 74 (1985) 269–272.

Itai, S., Nemoto, M., Kouchiwa, S., Murayama, H. and Nagai, T., Influence of wetting factors on the dissolution behaviour of flufenamic acid. *Chem. Pharm. Bull.*, 33 (1985) 5464–5473.

Mukerjee, P., Solubilization of benzoic acid derivatives by non-ionic surfactants. Location of solubilizates in hydrocarbon core of micelles and poly (oxyethylene) mantle. *J. Pharm. Sci.*, 60 (1971) 1528–1531.

Murthy, K.S. and Ghebre-Sellasie, I., Current Perspectives on the dissolution stability of solid oral dosage forms. *J. Pharm. Sci.*, 82 (1993) 113–125.

Neuvonen, P.J., Pentikainen, P.J. and Elfving, S.M., Factors affecting the bioavailability of phenytoin. *Int. J. Clin. Pharmacol.*, 15 (1977) 84–89.

Nishikido, N., Moroi, Y., Uehara, H. and Matuura, R., Effect of alcohols on the micelle formation of non-ionic surfactants in aqueous solutions. *Bull. Chem. Soc. Japan*, 47 (1974) 2634–2638.

Pandit, N.K., Strykowski, J.M., Mcnally, E.J. and Waldbillig, A.M., Surfactant solutions as media for dissolution testing of a poorly water-soluble drug. *Drug Dev. Ind. Pharm.*, 11 (1985) 1797–1818.

Parrot, E. and Sharma, V., Dissolution Kinetics of benzoic acid in high concentrations of surface-active agents. *J. Pharm. Sci.*, 56 (1967) 1341–1343.

Philip, J., Holcomb, I.J. and Fusari, S.A., Phenytion. In: Flory, K. (Ed.), *Analytical Profiles of Drug Substances*, vol. 13, Academic Press, Orlando, FL, 1984, pp. 417–445.

Rubino, J.T., Halterlein, L.M. and Blanchard, J., The effects of aging on the dissolution of phenytoin sodium capsule formulations. *Int. J. Pharm.*, 26 (1985) 165–174.

SAS/Stat User's Guide (Release 6.03), SAS Institute, Inc., Cary, North Carolina, 1988.

Serajuddin, T.A.M. and Jarowski, C.I., Influence of pH on release rate of phenytoin sodium from slow-release dosage forms. *J. Pharm. Sci.*, 82 (1993) 306–310.

Shah, V.P., Prasad, V.K., Alston, T., Cabana, B.E., Gural, R.P. and Meyer, M.C., Phenytoin I: In-vitro – in-vivo correlation for 100 mg phenytoin sodium capsules. *J. Pharm. Sci.* 72 (1983) 306–308.

Stavchansky, S. and Gowan, W.G., Evaluation of the bioavailability of a solid dispersion of phenytoin in polyethylene glycol 6000 and a commercial phenytoin sodium capsule in the dog. *J. Pharm. Sci.*, 73 (1984) 733–736.

Sved, S., Hossie, R.D., McGilvery, I.J., Beaudoin, N. and Brien, R., Bioavailability, Absorption and dissolution kinetics of phenytoin formulations. *Can. J. Pharm. Sci.*, 14 (1979) 67–71.

USP 23-NF 18, 1995, 12601 Twinbrook parkway, Rockville, MD 20852.

Walkling, W.D., Nayak, R.K., Plostnieks, J. and Cressman, W.A., A partially organic dissolution medium for griseofulvin dosage forms. *Drug Dev. Ind. Pharm.*, 5 (1979) 17–27.

Yakou, S., Yamazaki, S., Sonobe, T., Sugihara, M., Fakumuro, K. and Nagai, T., Particle size distribution affects the human bioavailability of phenytoin. *Chem. Pharm. Bull.*, 34 (1986) 4400–4402.