Study of Nimesulide Release from Solid Pharmaceutical Formulations in Tween 80 Solutions

STEFANA-OANA PURCARU(1), MIHAELA IONESCU(2), CRISTINA RANETI(2), VALENTINA ANUTA(3), I. MIRCIOIU(4), IONELA BELU(1)

(1) Faculty of Pharmacy, University of Medicine and Pharmacy of Craiova; (2) Army Center for Medical Research, Bucharest; (3) Biopharmacy & Pharmacol Res S.A., Bucharest; (4) Faculty of Pharmacy, “Carol Davila” University of Medicine and Pharmacy, Bucharest

ABSTRACT Nimesulide is a weakly acidic non-steroidal anti-inflammatory drug (NSAIDs). Like many non-steroidal anti-inflammatory drugs, Nimesulide is very sparingly soluble in water (≈ 0.01 mg/mL). The poor aqueous solubility and wettability of Nimesulide gives rise to difficulties in pharmaceutical formulations for oral or parenteral delivery, which may lead to variable bioavailability. Based on the Biopharmaceutical Classification System (BCS), Nimesulide is considered a BCS 2 drug (poorly soluble and highly permeable). Solubilization in surfactant solutions above critical micelle concentration (CMC) offers one approach to the formulation of poorly soluble drugs. Weakly acidic and basic drugs may be brought into solution by the solubilizing action of surfactants. In this study, different concentrations of Tween 80 was used in combination with buffer (pH 7.4) to increase the solubility of Nimesulide. The results show that the dependence of the released amount on the Tween concentration is not linear, very low Tween concentration showing a decrease of “solubility”, probably connected to a critical micelle concentration at the interface Nimesulide solution. An “analytical” artefact connected to a decreasing ultraviolet absorption of Nimesulide because of Nimesulide precipitation, the formation of a colloidal solution is possible, and the phenomenon remains to be searched further. It is hard to explain that for an almost complete solubilization a significant Tween quantity is necessary and this should be more than that of other slightly soluble drugs.

KEY WORDS Nimesulide, solubilization, Tween 80, critical micelle concentration (CMC)

Introduction

Oral dosage form represents the most common route for drug administration into the human body because it leads to a better patient compliance and it is very versatile for what concerns dosing conditions [1, 2]. Unfortunately, however, this strategy fails when dealing with low bioavailable drugs like those belonging to the widely employed anti-inflammatory class [3]. Although bioavailability, defined as the rate and extent to which the active drug is absorbed from a pharmaceutical form and becomes available at the site of drug action [4], depends on several factors, usually, drug solubility in an aqueous environment and drug permeability through lipophilic membranes play the role of key parameters [2]. In fact, only solubilized molecules can be absorbed by the cellular membranes to subsequently reach the site of drug action (vascular system for instance).

According to the high or low values assumed by these parameters, drugs can be divided in four different classes [5] and a drug can be defined bioavailable if it belongs to the first class (high solubility and permeability). Many different techniques are commonly used to improve the bioavailability of poorly water soluble but permeable drugs (second class [5]).

Drug bioavailability, in the case of oral administration, is also strongly affected by intestinal permeability. Therefore, drug permeation studies result of paramount importance for the development of those strategies aimed to improve drug absorption and the necessity of understanding the basic mechanisms ruling the drug transfer through the intestinal epithelium arises [6]. It was demonstrated [7] that in vivo drug permeation through the intestinal mucosa mainly takes place according to a passive diffusive mechanism whose rate determining step is represented by the cellular membrane crossing, while a little effect would be exerted by the aqueous stagnant layer arising at the intestinal wall [7]. Although, it is usually affirmed that lipophilic drugs follow a transcellular pathway in their intestinal membrane crossing, while hydrophilic ones undertake a paracellular pathway (they would diffuse through the water filling the intercellular voids), today the transcellular way is thought to be the main transport mechanism, both in rats and in human beings, regardless the drug physicochemical properties [8, 9].

Nimesulide, chemically 4’-nitro-2’-phenoxy methane sulfonanilide, is a weakly acidic
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(pKa 6.5) nonsteroidal anti-inflammatory drug (NSAIDs). It differs from other nonsteroidal anti-inflammatory drugs in that its chemical structure contains a sulfonanilide moiety as the acidic group rather than a carboxylic group (Figure 1).

![Figure 1 – Structure of Nimesulide.](image)

Nimesulide shows high anti-inflammatory, antipyretic, and analgesic activities in addition to low toxicity, a moderate incidence of gastric side effects, and a high therapeutic index [10]. Nimesulide is a relatively weak inhibitor of prostaglandin synthesis in vivo and appears to exert its effect through a variety of mechanisms including free radical scavenging, effects on histamine release, the neutrophil myeloperoxidase pathway, bradykinin activity, tumor necrosis factor-a release, cartilage degradation, metalloprotease synthesis, phosphodiesterase type IV inhibition, platelet aggregation, and synthesis of platelet activating factor. It also exhibits a significant selectivity toward cyclooxygenase-2 (COX-2) versus COX-1 inhibition, which may explain the lower incidence of gastric side effects. However, recent findings reported that Nimesulide has a higher risk of hepatic toxicity when compared to other marketed NSAIDs [11, 12]. Like many non-steroidal anti-inflammatory drugs, Nimesulide is very sparingly soluble in water (~0.01 mg/mL) [13]. The poor aqueous solubility and wettability of Nimesulide gives rise to difficulties in pharmaceutical formulations for oral or parenteral delivery, which may lead to variable bioavailability [14].

Based on the Biopharmaceutical Classification System (BCS), Nimesulide is considered a BCS 2 drug (poorly soluble and highly permeable) [15] therefore, dissolution is a limiting step for its absorption [16].

Solubilization in surfactant solutions above critical micelle concentration (CMC) offers one approach to the formulation of poorly soluble drugs [17]. Weakly acidic and basic drugs may be brought into solution by the solubilizing action of surfactants [18].

In this study, different concentrations of a non-ionic surfactant Polysorbate 80 (Tween 80, CMC 0.012 mM) was used in combination with buffer (pH 7.4) to increase the solubility of Nimesulide.

**Material and Methods**

**Dissolution methods**

The dissolution profiles were studied using USP apparatus 2, in 900 mL of dissolution media: USP phosphate buffers of pH 7.4 with 2.5%, 1%, 0.5%, 0.1%, 0.05% and 0.01% Tween 80, USP acetate buffers of pH 4.5. Stirring rates were 100 rpm, with a constant temperature bath at 37±0.5°C. Four-millilitre samples were drawn at 5, 10, 15, 20 30, 45 and 60 minutes and replenished with 4 mL of fresh dissolution medium. Analytical assay method was spectrophotometric, determinations being performed at 274 nm.

The calibration curve (four standards) is made in phosphate buffer pH 7.4 with 2.5% Tween 80. In all other media, Nimesulide has not dissolved. All readings have been done with the same calibration.

For each dissolution three liters of buffer with the specific percentage of Tween 80 have been prepared, after which 900 mL have been introduced in each vessel; the remaining of the 300 mL solution has been used for blank and standards.

**Results and Discussion**

**The release in 2.5% Tween 80**

The release has been very rapid, without "time-lag". However, the release has not been complete, in 60 minutes the released quantity being under 92%. The value does not seem to be a saturation value, a more complete release being possible after an hour (Figure 2).

**The release in 1% Tween 80**

The release is instantaneous and the concentration is constant in a 10-60 minute interval, without any evidence that it might increase afterwards. Apparently, the release is limited by solubility, which seems to be of 78–79% in 1% Tween 80 (Figure 3).

**The release in 0.5% Tween 80**

The release is immediate. It seems that the value of 77% is a saturation value. It seems to be a little bit less than the saturation value in 1% Tween 80 (Figure 4).

**The release in 0.10% Tween 80**

The release is immediate. It seems that the value of 52% is more like an error in noticing number 1 in number 2 vessel. The value of 49% appears as a saturation value (Figure 5).
The release in 0.50% Tween 80
The release is immediate. The jump from 15 to 20 minutes is less explainable. The saturation value seems to be 57% (Figure 6).

Figure 2 – Release profile in 2.5% Tween 80.

Figure 5 – Release profile in 0.10% Tween 80.

Figure 6 – Release profile in 0.05% Tween 80.

The release in 0.01% Tween 80
The looks of the curve is that of a saturation curve, probably the value of 51–52% (Figure 7).

Figure 3 – Release profile in 1% Tween 80.

Figure 7 – Release profile in 0.01% Tween 80.

The comparison of the dissolution profiles
The representation of all the dissolution curves confirms “the saturation curve” character for all the concentrations less that of 2.5%. In these conditions, the study appears as an estimation of the dependence of Nimesulide “solubility” as function of the Tween concentration (Figure 8).

The representation of the “solubility” related to the Tween concentration reveals at least strange behaviour. The solubility seems to decrease from...
the concentration of 0.05% to 0.1% and then increases with the Tween concentration (Figure 9). This could be an analytical “misinterpretation”. Standard curves were in fact straight lines with a very good correlation coefficient in all cases. However, for small Tween concentration, the solutions were in fact opalescent and there could be a systematic error in estimation (“bias”) of the whole data set. Unfortunately, there are not accurate estimations of concentrations in opalescent solutions. In the range of used Tween concentrations, its critical micellar concentration (CMC) is included. We do not know which is the value of CMC in phosphate buffer and in Nimesulide presence.

Since solutions used in the experiment were prepared by measuring a Tween volume, not by weighing, CMC should be anyway not too far from the value 0.01%. Consequently, discontinuity or even inversion of the effect could be a critical behaviour around CMC.

In fact, the surfactant accumulates on the interface of Nimesulide particles – solution and in that interface, all phenomena and all concentrations significantly differ from those in bulk of solution. Any imagined mechanism concerning interface phenomena, where electrical structures appear also, remains an unverifiable assumption.

In the end, we should keep this “abnormal” behaviour, as “normal” in the neighbourhood of cmc.

The release model

The only case in which the release appeared really as a “release curve” was the release in 2.5% Tween. A representation of the released quantity as function of square root of time was made in order to test a possible release following Higuchi law. If this were the case, the experimental points should range on a line, but it was not the case (Figure 10).

For the 2.5% case, we have gone further and made a representation of ln (1-Rexp/100) depending on time (Noayes–Whitney “linearized” law). Neither by this way a linear dependence has been obtained (Figure 11).
Figure 11 – Representation of ln(1-Rexp/100) depending on time.

However, the curve appears to be a release curve. To correlate the data we have chosen the Weibull empirical law:

\[ R(\%) = 100 \left(1 - e^{-at^\beta}\right) \]

\[ e^{-at^\beta} = 1 - R/100 \]

\[ \ln\ln\left(\frac{1}{100}\right) = \ln\alpha + \beta\ln(t) \] (Fr1)

The \(\ln(1-R/100)\) representation depending on \(\ln t\) has really led to a line as it may be seen in the bellow Figure 12.

Figure 12 – Representation of \(\ln(1-R/100)\) depending on \(\ln t\).

Conclusions

1. The Nimesulide release from tablets at pH 7.4 is an immediate release, the released amount being apparently limited by solubility, a limit that depends on the Tween concentration.

2. The dependence of the released amount on the Tween concentration is not linear, very low Tween concentration showing also a decrease of “solubility”, probably connected to a critical micelle concentration at the Nimesulide solution interface. An “analytical” artefact connected to a decreasing ultraviolet absorption of Nimesulide because of Nimesulide precipitation, the formation of a colloidal solution is possible, and the phenomenon remains to be searched further.

3. It is hard to explain why for an almost complete solubilization is necessary a significant higher Tween quantity than for other slightly soluble drugs.

4. The model followed by the kinetic release is Weibull, but the initial release could be modelled by Higuchi square root law also.

References

1. Waterbeemd van de H. – (2000) Intestinal permeability: prediction from theory. In: Dressman JB, Lennernäs H (eds), Oral drug absorption, prediction and assessment, Marcel Dekker, New York, 31–50.

2. Yu LX, Gatlin L, Amidon GL – (2000) Predicting oral drug absorption. In: Amidon GL, Lee PI, Topp EM (eds), Transport processes in pharmaceutical systems, Marcel Dekker, New York, 377–409.

3. Grassi M, Coceani N, Magarotto L – (2002) Modelling partitioning of sparingly soluble drugs in a two-phase liquid system, Int J Pharm, 239(1–2):157–169.

4. Pharmacos 4 – (2009) Eudralex collection, medicinal products for human use: guidelines, volume 3C, 234 (website: http://pharmacos.eudra.org/F2/eudralex/vol-3/home.htm).

5. Amidon GL, Lennernäs H, Shah VP, Crison JR – (1995) A theoretical basis for a biopharmaceutic drug classification: the correlation of in vitro drug product dissolution and in vivo bioavailability, Pharm Res, 12(3):413–420.

6. Grassi M, Cadelli G – (2001) Theoretical considerations on the in vivo intestinal permeability determination by means of the single pass and recirculating techniques, Int J Pharm, 229(1–2):95–105.

7. Fagerholm U, Lennernäs H – (1995) Experimental estimation of the effective unstirred water layer thickness in the human jejunum, and its importance in oral drug absorption, Eur J Pharm Sci, 3(5):247–253.

8. Fagerholm U, Johansson M, Lennernäs H – (1996) Comparison between permeability coefficients in rat and human jejunum, Pharm Res, 13(9):1336–1342.

9. Meriani F, Coceani N, Sirotti C, Voinovich D, Grassi M – (2004) In vitro Nimesulide absorption from different formulations, J Pharm Sci, 93(3):540–552.

10. Davis N, Brogden RN – (1994) Nimesulide: an update of its pharmacodynamic and pharmacokinetic properties and its therapeutic efficacy, Drugs, 48(3):431–454.

11. Macia MA, Carvajal A, Del Pozo JG, Vera E, Del Pino A – (2002) Hepatotoxicity associated with Nimesulide: data from the Spanish Pharmacovigilance System, Clin Pharmacol Ther, 72(5):596–597.

12. Rodrigo L, De Francisco R, Pérez-Pariente JM, Cadahia V, Tojo R, Rodríguez M, Lucena MI, Andrade RJ – (2002) Nimesulide-induced severe hemolytic anemia and acute liver failure leading to liver transplantation, Scand J Gastroenterol, 37(11):1341–1343.

13. Piel G, Pirotte B, Delneuville I, Neven P, Llabres G, Delarge J, Delattre L. – (1997) Study of the influence of both cyclodextrins and L-Lysine on the aqueous solubility of nimesulide: isolation and characterization of nimesulide-L-Lysine-cyclodextrin complexes, J Pharm Sci, 86(4):475–480.

14. Nalluri Buchi N, Chowdar KPR, Murthy KVR, Hayman AR, Becket G – (2003) Physicochemical characterization and dissolution properties of nimesulide-cyclodextrin binary systems, AAPS PharmSciTech, 4(1):E2.

15. Abdelkader H, Abdallah OY, Salem HS – (2007) Comparison of the effect of tromethamine and polyvinylpyrrolidone on dissolution properties and analgesic effect of Nimesulide, AAPS Pharm Sci Tech, 8(3):E1–E8.

16. Carrasco-Portugal MC, Flores-Murrieta FJ – (2008) Comparison of suspension composition on the pharmacokinetics of Nimesulide in rats, Proc West Pharmacol Soc, 51:58–59.

17. Attwood D, Florence AT – (1983) Surfactant system, Chapman and Hall, New York.
18. Martin A, Bustamante P, Chun AHC – (1993) Solubility and distribution phenomena. In: ***, Physical phenomena, 4th edition, Lea & Febiger, Philadelphia, 212–250.

19. *** – (2009) www.sigmaaldrich.com.

20. Malik WU, Jhamb OP – (1970) Critical micelle concentration of some polyoxyethylated non-ionic surfactants and the effect of additives, Colloid Polymer Sci, 242(1–2):1209–1211.

21. Zhang H, Yao M, Morrison RA, Chong S – (2003) Commonly used surfactant, Tween 80, improves absorption of P-glycoprotein substrate, Digoxin, in rats, Arch Pharm Res, 26(9):768–772.

Correspondence Address: Professor Constantin Mircioiu, Pharm., PhD, Department of Biopharmacy, Faculty of Pharmacy, “Carol Davila” University of Medicine and Pharmacy, 6 Traian Vuia Street, 020956 Bucharest, Romania; Phone +4021–318 07 50, e-mail: constantin.mircioiu@yahoo.com