International Journal of Pharmaceutics 428 (2012) 118–120

Contents lists available at SciVerse ScienceDirect

International Journal of Pharmaceutics

journal homepage: www.elsevier.com/locate/ijpharm

Rapid communication

In situ controlled crystallization as a tool to improve the dissolution of Glibenclamide

Amal Ali Elkordy[a,*], Ayobami Jatto[a], Ebtessam Essa[b,c]

[a] University of Sunderland, Department of Pharmacy, Health and Well-being, Sunderland, SR1 3SD, UK
[b] University of Umm Al Qura, Faculty of Pharmacy, Department of Pharmaceutics, Makkah, Saudi Arabia
[c] University of Tanta, Faculty of Pharmacy, Department of Pharmaceutical Technology, Tanta, Egypt

ARTICLE INFO

Article history:
Received 9 January 2012
Received in revised form 29 February 2012
Accepted 29 February 2012
Available online 7 March 2012

Keywords:
Controlled crystallization
Glibenclamide
Cremophor RH40
Solutol HS-15

ABSTRACT

For pharmaceutical purpose, micro-sized drugs are needed for many delivery systems, such as pulmonary and oral drug delivery systems. Many strategies have been employed to reduce the particle size of poorly water soluble drugs. Microcrystals could be produced by controlled association of drug in order to obtain naturally grown particles. The aim of this work was to increase the aqueous solubility and dissolution of Glibenclamide. The in situ controlled crystallization process was conducted in the presence of the non-ionic surfactants, Cremophor RH40 and Solutol HS-15 (0.75 and 1.5%, w/v), as protective stabilizing agents against agglomeration. In addition, these surfactants inhibit P-glycoprotein that reduces intestinal absorption of Glibenclamide by efflux transportation. Crystal shape was changed and particle size was reduced by about 15-folds, compared to control untreated drug. Differential Scanning Calorimetry (DSC) results indicated no interaction between the drug and the stabilizer. Microcrystals showed marked increase in the drug dissolution, Solutol HS-15 at 1.5% (w/v) concentration showing the highest dissolution efficiency. It could be concluded that in situ controlled crystallization using surfactants are promising method to improve dissolution of Glibeclamide as a model poorly water soluble drug.

The solubility of a drug is an important factor in determining the rate and extent of its absorption. Many drugs are poorly soluble or insoluble in water, which results in poor bioavailability. One way to improve the dissolution rate is to reduce particle size. For pharmaceutical purposes, micron-sized drugs are required for several dosage forms—such as for oral or pulmonary use. The common way for micronization is the milling of previously formed larger crystals. However, this technique is ineffective and shows disadvantages such as electrostatic effects, broad particle size distributions, and the tendency of the particles to grow (Rasenack et al., 2004). In situ microcrystallization is a suitable method for the production of micron-sized drugs and overcome the problem of milling (Rasenack and Muller, 2002; Kim et al., 2003). During the in situ crystal formation, a new surface is formed that makes the system thermodynamically unstable. A stabilizer which has affinity to the crystal surface can cover the newly formed surfaces and in turn might reduce the surface energy, producing micron-sized particles stabilized against crystal growth by steric hindrance (Rasenack et al., 2004).

Glibenclamide (GLB) is an oral hypoglycemic agent used in the treatment of type II diabetes. Being a Class II drug, the rate of drug dissolution is almost certainly the principal limitation to its oral absorption. Consequently, improving the aqueous solubility of the lipophilic GLB can improve its clinical performance, and eventually decrease the dose (Wei et al., 2006). Many approaches have been applied for the enhancement of the solubility of GLB, for example; complex formation with cyclodextrins (Esclusa-Díaz et al., 1994), solid dispersion systems (Valleri et al., 2004), and crystalline-form conversion (Hassan et al., 1997).

The aim of this work was to improve GLB solubility and dissolution by in situ controlled crystallization using the solvent exchange technique (Varshosaz et al., 2008). The selected de-agglomerating (stabilizer) agents were Solutol HS-15 (So) and Cremophor RH40 (Cr), at two different concentrations 0.75% (w/v) (So1 and Cr1) and 1.5% (w/v) (So2 and Cr2). These non-ionic surfactants were particularly selected based on their known inhibition effect to P-glycoprotein (Coon et al., 1991; Rege et al., 2002; Benoit and Lanprecht, 2004; Peltier et al., 2006). This P-glycoprotein reduces GLB intestinal absorption by efflux transportation, so reduces its plasma concentration (Srirangam and Vidya, 2010). Consequently, any increase in bioavailability would be due to the dual effect of both size and stabilizer.

The in situ microcrystals were prepared by instantaneous mixing two liquids containing Solutol HS-15 or Cremophor RH40 (BASF Corporation, Germany) in aqueous solution and drug in an organic solvent. Accurately weighted 1.0 g GLB (Sigma–Aldrich, UK) was

* Corresponding author. Tel.: +44 01915152576; fax: +44 01915153405.
E-mail address: amal.elkordy@sunderland.ac.uk (A.A. Elkordy).

0378-5173/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
doi:10.1016/j.ijpharm.2012.02.046

Table 1
Particle size (μm), polydispersity index (PDI), saturated aqueous solubility (SAS, μg/mL), percentage drug released after 5 min (Q_5) and percentage dissolution efficiency (%DE) of Glibenclamide from different microcrystal formulations and control.

Formula[a]	Stabilizer concentration (%)	Particle size (μm)	PDI	SAS (μg/mL)	Q_5 (%)	%DE
Control	–	22.3 ± 2.3	0.66 ± 0.23	15.34 ± 2.8	19.0 ± 2.6	50.5 ± 4.4
MCD	–	4.72 ± 0.83	0.25 ± 0.06	48.21 ± 7.1	44.2 ± 3.1	72.4 ± 2.7
Cr1	0.75	2.21 ± 0.31	0.21 ± 0.04	106.2 ± 6.3	66.1 ± 2.9	84.6 ± 4.8
Cr2	1.5	2.12 ± 0.12	0.15 ± 0.05	187.2 ± 5.9	74.2 ± 4.2	90.0 ± 3.9
So1	0.75	2.70 ± 0.65	0.18 ± 0.06	115.6 ± 3.2	72.6 ± 2.4	89.6 ± 5.2
So2	1.5	1.52 ± 0.43	0.12 ± 0.02	119.1 ± 5.8	92.5 ± 3.7	97.7 ± 2.3

All experiments were conducted in triplicates.
[a] Control for commercial drug. MCD is the microcrystalline drug without stabilizer. Cr1, Cr2, So1, So2 are microcrystals with stabilizers 0.75% Cremophor RH40, 1.5% Cremophor RH40, 0.75% Solutol HS-15 and 1.5% Solutol HS-15, respectively.

dissolved in 30 mL of dichloromethane (as the solvent) and 1.0 or 2.0 g of stabilizing agent in 100 mL of water (as non-solvent). By batch-wise mixing during stirring for about 10 min, microcrystals are formed. The obtained crystals were recovered by centrifugation and drying. By changing surfactants' concentrations in the total crystallization solution, four formulations (Cr1, Cr2, So1 and So2) were obtained (Table 1), in addition to microcrystallized drug without stabilizer, MCD. Commercial drug was used as a control.

Scanning electron microscope (Hitachi, UK) images indicated that the crystal shape changed from rod-like for the untreated drug to diamond or cube shapes for the newly formed microcrystals in the presence of surfactants (Fig. 1). The particle size (Zeta Sizer Analyzer, New York) for control was about 22.3 ± 2.6 μm with high polydispersity index (the width of the particle size distribution curve).

With microcrystals, particle size was significantly ($P < 0.05$, using Student's *t*-test) reduced for all formulations. Such reduction was by about 5-folds for MCD up to 15-folds in Cr2, relative to control (Table 1), with polydispersity index less than 0.2, thus indicating distribution homogeneity.

Accordingly, surfactants can impede the molecular association and the crystal growth by forming a protective layer around crystals.

Saturated drug solubility for all microcrystalline formulations was tested by rotating excess amount of each sample with 10 mL distilled water for 72 h. Filtered supernatants were assayed spectrophotometrically at 300 nm. MCD increased drug solubility by about 3-folds (Table 1). Microcrystals with both stabilizers further increased solubility, with Cr2 showing the highest solubility. Increasing Cremophor RH40 concentration from 0.75 (Cr1) to 1.5% (w/v) (Cr2) increased drug solubility, while for Solutol HS-15 (So) there was no significant difference (between So1 and So2 ($P > 0.05$)) in solubility with increasing concentration (refer to Table 1).

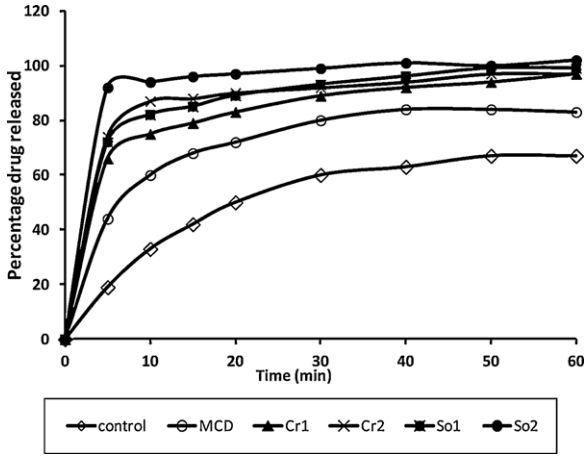

Fig. 2. Percentage drug release versus time plots of control untreated drug, drug-alone microcrystals (MCD), and microcrystals containing stabilizer [0.75% Cremophor RH40 (Cr1), 1.5% Cremophor RH40 (Cr2), 0.75% Solutol HS-15 (So1) and 1.5% Solutol HS-15 (So2)]. Standard error bars were omitted for clarity.

To estimate the effect of size reduction on the in vivo performance of GLB, it was important to study, under in vitro conditions, the release of the drug from different formulations. Dissolution studies were performed assuring sink conditions using USP apparatus II (Erweka, UK). Samples equivalent to 12 mg GLB were placed in 900 mL distilled water at 37 ± 0.5 °C, and stirred at 100 rpm. The releases of GLB from the tested formulations were plotted as percent amount of GLB released versus time (Fig. 2).

The percentage drug release after 5 min (Q_5) and dissolution efficiency, DE (calculated from the area under the curve at time t and measured using the trapezoidal rule and expressed as a percentage

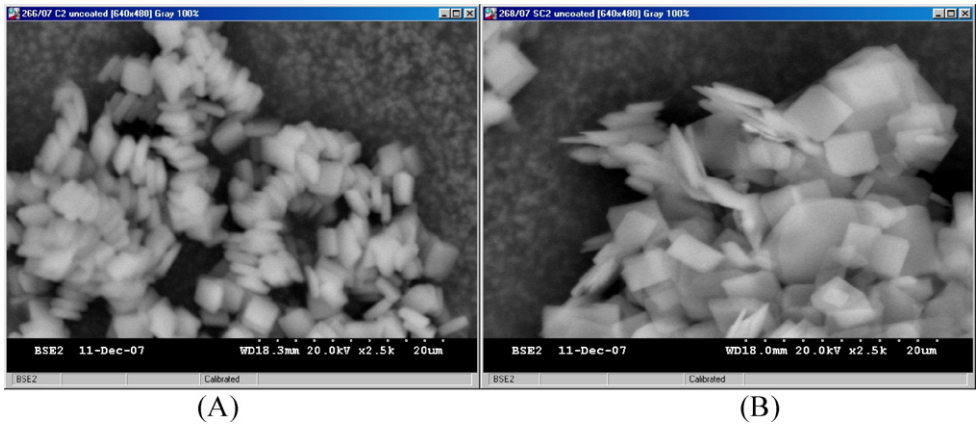

Fig. 1. Representative scanning electron microscope of Glibenclamide microcrystals in the presence of: (A) 1.5% (w/v) Cremophor RH40 and (B) 1.5% w/v Solutol HS-15.

of the area of the rectangle described by 100% dissolution in the same time (Khan, 1975)) were determined (Table 1). Regarding Q_5, all microcrystals showed a prompt drug release compared to control ($P < 0.05$), with So2 showing the highest release. All microcrystals significantly increased the DE ($P < 0.05$) compared to the commercial drug. Again, So2 showed the highest DE. Although both surfactants have similar HLB value (14–16), Solutol, HS-15 was superior in improving drug performance. The results could be explained by the reduced particle size. Additionally, the presence of protective stabilizer would have prevented crystal agglomeration in contact with water. Meantime, being adsorbed onto the surface of the drug particles during the phase separation step, it is expected that the surfactant would lower the contact angle between the hydrophobic drug particles and the aqueous medium, by reducing the interfacial tension, thus increasing drug wettability and consequently drug dissolution. DSC for control drug showed a sharp endothermic peak at 175 °C, T_m, corresponding to GLB melting. For So2, there was a slight shift to lower T_m of about 174 °C indicating the absence of any significant interaction between the drug and the stabilizer, implying that the size reduction and shape uniformity in microcrystals and also using of the non-ionic surfactants are the reasons for such improvement in drug dissolution. The overall results signify the usefulness of the in situ controlled crystallization with P-glycoprotein inhibitors in enhancing the oral bioavailability of GLB which could be estimated by the above results. Additionally, GLB formulation into inhalation dosage form is also a possibility.

References

Benoit, J.P., Lanprecht, L., 2004. Use of p-glycoprotein inhibitor surfactant at the surface of a colloidal carrier. Patent W02004071498.

Coon, J.S., Knudson, W., Clodfelter, K., Lu, B., Weinstein, R.S., 1991. Solutol HS 15, nontoxic polyoxyethylene esters of 12-hydroxystearic acid, reverses multidrug resistance. Cancer Res. 51, 897–902.

Esclusa-Díaz, M.T., Torres-Labandeira, J.J., Kata, M., Vila-Jato, J.L., 1994. Inclusion complexation of glibenclamide with 2-hydroxypropyl-β-cyclodextrin in solution and in solid state. Eur. J. Pharm. Sci. 16, 291–296.

Hassan, M.A., Sheikh Salem, M., Sallam, E., Al-Hindawi, M.K., 1997. Preparation and characterization of a new polymorphic form and a solvate of glibenclamide. Acta Pharm. Hung. 672, 81–88.

Khan, K.A., 1975. The concept of dissolution efficiency. J. Pharm. Pharmacol. 28, 48–49.

Kim, S.T., Kwon, J.H., Lee, J.J., Kim, C.W., 2003. Microcrystallization of indomethacin using a pH-shift method. Int. J. Pharm. 263, 141–150.

Peltier, et al., 2006. Enhanced oral paclitaxel bioavailability after administration of paclitaxel-loaded lipid nanocapsules. Pharm. Res. 33, 1243–1249.

Rasenack, N., Muller, B.W., 2002. Dissolution rate enhancement by *in situ* micronization of poorly water-soluble drugs. Pharm. Res. 19, 1894–1900.

Rasenack, N., Steckel, H., Müller, B.W., 2004. Preparation of microcrystals by in situ micronization. Powder Technol. 143–144, 291–296.

Rege, B.D., Kao, J.P., Polli, J.E., 2002. Effects of nonionic surfactants on membrane transporters in Caco-2 cell monolayers. Eur. J. Pharm. Sci. 16, 237–246.

Srirangam, P., Vidya, S.J., 2010. Modulation of the P-glycoproein-mediated intestinal secretion of Glibenclamide: *in vitro* and *in vivo* assessments. J. Young Pharm. 2, 379–383.

Valleri, M., Mura, P., Maestrelli, F., Cirri, M., Ballerini, R., 2004. Development and evaluation of glyburide fast dissolving tablets using solid dispersion technique. Drug Dev. Ind. Pharm. 305, 525–534.

Varshosaz, J., Talari, R., Mostafavi, A., Nokhodchi, A., 2008. Dissolution enhancement of gliclazide using *in situ* micronization by solvent change method. Powder Technol. 187, 222–230.

Wei, H., Lacan, R., Cirri, M., 2006. Biorelevant dissolution media as a predictive tool for glyburide a class II drug. Eur. J. Pharm. Sci. 19, 45–62.