Journal of Drug Delivery Science and Technology 41 (2017) 303–309

Contents lists available at ScienceDirect

Journal of Drug Delivery Science and Technology

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

Peceosomes for oral delivery of glibenclamide: In vitro in situ correlation

Amal A. Sultan, Sanaa A. El-Gizawy, Mohamed A. Osman, Gamal M. El Maghraby*

Department of Pharmaceutical Technology, College of Pharmacy, University of Tanta, Tanta, Egypt

ARTICLE INFO

Article history:
Received 5 May 2017
Received in revised form
28 July 2017
Accepted 8 August 2017
Available online 9 August 2017

Keywords:
Glibenclamide
Peceosomes
Drug release
In situ intestinal absorption
Correlation

ABSTRACT

Peceol containing niosomes (peceosomes) have been shown to widen absorption window of acidic drugs. However, the relationship between drug release and intestinal absorption is not clear. Accordingly, the objective was to probe peceosomes for enhancing intestinal absorption of glibenclamide with the goal of correlating drug release with intestinal absorption. Glibenclamide was encapsulated into peceosomes. The drug entrapment and release were determined at pH values of 6.6 and 7.4. In situ rabbit intestinal absorption of glibenclamide was monitored from peceosomes with reference to aqueous solution. The entrapment efficiency of glibenclamide was 89.3% and 68.7% with the release efficiency being 6.5% and 19.1% at pH 6.6 and 7.4, respectively. In situ perfusion of glibenclamide solution reflected incomplete absorption from duodenum and jejuno-ileum. Peceosomal encapsulation enhanced glibenclamide intestinal absorption with complete absorption being achieved from jejuno-ileum. Correlating intestinal absorption with in vitro release, the former was always greater. This indicates that drug release rate is not the rate limiting step. Taking this into consideration together with augmented jejuno-ileal transcellular absorption, intact peceosomal translymphatic absorption can be suggested. The study confirmed the ability of peceosomes to widen absorption window of acidic drugs and highlighted the need for drug retention in vesicles for efficient delivery.

1. Introduction

Glibenclamide is a potent sulphonyl urea oral hypoglycemic drug which stimulates pancreatic beta cells to liberate insulin. The drug was also reported to have a potential to reduce the insulin clearance by the liver to maintain high insulin blood level [1,2]. Glibenclamide is classified as a poorly soluble highly permeable drug [3,4]. This results in low and variable bioavailability of glibenclamide after oral administration [5]. Moreover, glibenclamide is a weak acidic drug with a pKa of 5.3. This makes it susceptible for pH dependent solubility, ionization and intestinal absorption with a possibility for narrow absorption window. The absorption rate of this drug is thus expected to decrease from the distal regions of the gastrointestinal tract (GIT). This site dependent absorption can contribute further to the variability and reduction in oral bioavailability of glibenclamide [1]. Glibenclamide absorption from different gastrointestinal sites was previously investigated and the results confirmed the pH partition hypothesis [6]. Therefore, development of a delivery system which can deliver the drug through the GIT irrespective to the pH of the absorption site can be of great benefit. Niosomes are vesicular nanostructures which were able to enhance oral bioavailability of lipophilic drugs [7,8]. These vesicular systems are composed mainly of nonionic surfactants and cholesterol as the basic components. Although structurally similar to liposomes, they can overcome liposomal problems such as difficulty of scaling up and high cost [9]. Peceol containing niosomes have been shown to widen the absorption window of another acidic drug [10]. This study employed in situ rabbit intestinal perfusion technique and monitored the amount of drug disappearing from the intestine with no consideration for the effect of drug release from such vesicular carriers on the rate of drug absorption. This work employed peceol as a main component of niosomal vesicles based on its role as a potential membrane permeability enhancer [11]. Accordingly, the aim of the present research was to investigate the potential of peceol incorporating

* Corresponding author.
E-mail addresses: amal_aahmed2009@yahoo.com (A.A. Sultan), selgizawy@hotmail.com (S.A. El-Gizawy), mosman4444@yahoo.com (M.A. Osman), gmmelmag@yahoo.com (G.M. El Maghraby).

http://dx.doi.org/10.1016/j.jddst.2017.08.003
1773-2247/

niosomes "peceosomes" as a vesicular carrier for enhancing intestinal absorption of glibenclamide. The work was extended to correlate the in vitro release data to in situ rabbit intestinal perfusion results.

2. Materials and methods

2.1. Materials

Glibenclamide was obtained as a gift sample from Sigma for Pharmaceutical Industries, Quesna, Egypt. Glyceryl monooleate (peceol) was procured as a gift from Gattefosse, Saint-Priest Cedex, France. Acetonitrile (HPLC specifications) was from SDFCL (S D Fine Chem Limited), Mumbai, India. Sorbitan monostearate (Span 60) was purchased from Sigma Chemical Co. (St. Louis, MO, USA). Cholesterol, ethanol, potassium dihydrogen phosphate, potassium chloride, sodium chloride and disodium hydrogen phosphate were procured from El-Nasr Pharmaceutical Chemicals Company, Cairo, Egypt.

2.2. Chromatography

Glibenclamide was quantified employing high pressure liquid chromatographic (HPLC) system (Agilent technologies 1260 infinity, DE, Germany). Chromatographic separation was accomplished utilizing an ODS reversed phase column of 4.6 mm internal diameter, 150 mm length and 5 μm average particle size (GL Sciences Inc., Japan). A mobile phase comprising a mixture of 70% acetonitrile and 30% potassium dihydrogen phosphate solution (25 mM, adjusted to pH3.5 with *ortho*-phosphoric acid) was pumped isocratically at a flow rate of 1 ml min^{-1}. Samples of 30 μl volume were automatically injected via an automatic sampling system (TCC 1260) and the column effluent was monitored at 228 nm using a variable wavelength UV detector (VWD 1260). The whole chromatographic system was under computer control with the data analysis being conducted utilizing Agilent OpenLAB ChemStation software. The method was validated with regard to the International conference on harmonization (ICH) guidelines.

2.3. Preparation of peceosomes

The technique employed to prepare peceosomes was previously elucidated in the literature [10]. Concisely, Peceosomes components comprising span 60, cholesterol and peceol (1.2, 0.3 and 0.3 g, respectively) were dispersed in 1.5 g of ethanol before being heated to 65±1 °C until clarity. Glibenclamide (1.5 mg) was dissolved in the developed ethanolic mixture followed by addition of 1.5 ml of water with gentle mixing while kept warm till homogeneity. The mixture is then allowed to cool down with the mixing being maintained to produce peceosomes proconcentrate. This proconcentrate was hydrated gradually with water to yield 50 ml crude peceosomal dispersion that was left then overnight for complete hydration at ambient temperature. The crude peceosomes were diluted with phosphate buffered saline (PBS; 1:3 v/v) and bath sonicated for 30 min instantly prior to either in vitro evaluation or in situ intestinal perfusion study.

2.4. Particle size analysis

Peceosomes were appropriately diluted with filtered water and then sonicated for few minutes to create uniformly dispersed vesicles free from vesicular clumps. The mean vesicular size and polydispersity index (PDI) were measured utilizing a Zetasizer Nano-ZS dynamic light scattering (DLS) instrument (Malvern Instruments Ltd., Worcestershire, UK). The size analysis was conducted at 25 °C with the scattering angle being 90°. The recorded size data were obtained from 16 measurements and presented as the mean (Z-average) ± the standard deviation (SD) employing the instrumental software.

2.5. Entrapment efficiency determination

Drug entrapment efficiency in a vesicular system was expressed as the percentage fraction entrapped of the initially added drug. Consequently, entrapment efficiency of glibenclamide in peceosomes was considered after separation of the unentrapped glibenclamide by dialysis. Instantly after hydration of the crude formulation with PBS, peceosomes dispersion (5 ml) was incubated into dialysis sacs (Cellulose dialysis tubing, Serva, Germany) and dialysed for 4 h against 100 ml of PBS. Glibenclamide concentration in the dialysate was measured by HPLC and was corrected for dialysate volume to measure the amount of unentrapped drug. The entrapment efficiency percentage of glibenclamide was mathematically computed utilizing the following equation [12,13]:

$$''\text{Entrapment efficiency } (\%) = [(\text{Ct} - \text{Cf})/\ \text{Ct}] \times 100''$$

where C_t is the total amount of the drug added to 5 ml peceosomes and C_f is the amount of the free un-entrapped drug.

2.6. In vitro glibenclamide release study

In vitro release of glibenclamide from peceosomes was investigated utilizing dialysis method. The developed crude peceosomes were hydrated and sonicated for 30 min immediately before being dialysed against PBS adjusted to pH values of 6.6 and 7.4. This was conducted to mimic the vesicles used in the in situ intestinal perfusion conditions. Cellulose dialysis tubing (Serva, Germany) was soaked overnight in distilled water before use for the release study. This was performed to confirm complete swelling of the membrane to provide constant pore width throughout the experiment. Dialysis sacs were suitably cut and filled with 5 ml of the diluted peceosomes and immersed in a 50 ml release medium being maintained at 37 °C with a constant mild stirring rate. Samples (2 ml) of the release medium were withdrawn at predefined time points for 2 h and replenished with equivalent volume of the fresh medium (preheated to 37 °C). Glibenclamide content in the collected samples was analyzed using HPLC. The release tests were performed in triplicates and release profiles were constructed as plots of the cumulative amount released as a function of time. These profiles were utilized to calculate the release efficiency from the area under the release profile at time (t) employing the nonlinear trapezoidal rule. The release efficiency was articulated as a percentage of the area under the rectangle formed assuming 100% release in the same time [14]. The difference between the release profiles was assessed using the similarity factor test which was computed using the following equation:

$$F2 = 50 \cdot \log\left\{\left[1 + \frac{1}{n}\sum_{t=1}^{n}(R_t - T_t)^2\right]^{-0.5}\right\} \cdot 100$$

“where n represents the number of data points, R_t and T_t correspond to the amounts of the drug released from peceosomes at the two pH values at time t”.

2.7. In situ intestinal absorption studies

The study protocol and manipulation of the test animals were