European Journal of Pharmaceutical Sciences 92 (2016) 173–182

Contents lists available at ScienceDirect

European Journal of Pharmaceutical Sciences

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

Pharmacokinetic considerations of nanodelivery to the brain: Using modeling and simulations to predict the outcome of liposomal formulations

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ARTICLE INFO

Article history:
Received 5 April 2016
Received in revised form 30 June 2016
Accepted 3 July 2016
Available online 5 July 2016

Keywords:
Liposomes
Nanocarriers
Brain delivery
Blood–brain barrier
Release rate
Modeling and simulation

ABSTRACT

The use of nanocarriers is an intriguing solution to increase the brain delivery of novel therapeutics. The aim of this paper was to use pharmacokinetic analysis and simulations to identify key factors that determine the effective drug concentration–time profile at the target site in the brain. Model building and simulations were based on experimental data obtained from the administration of the opioid peptide DAMGO in glutathione tagged PEGylated liposomes to rats. Different pharmacokinetic models were investigated to explore the mechanisms of increased brain delivery. Concentration–time profiles for a set of formulations with varying compound and carrier characteristics were simulated. By controlling the release rate from the liposome, the time profile and the extent of brain delivery can be regulated. The modeling did not support a mechanism of the liposomes passing the brain endothelial cell membrane in an intact form through endocytosis or transcytosis. The most likely process was found to be fusion of the liposome with the endothelial luminal membrane. The simulations revealed that low permeable compounds, independent on efflux, will gain the most from a nanocarrier formulation. The present model based approach is useful to explore and predict possibilities and limitations of carrier-based systems to the brain.

1. Introduction

There is an emerging need for effective treatments for central nervous system (CNS) disorders (Nuttand and Attridge, 2014). The use of nanocarriers offers an extended opportunity to attain the desired CNS effects of these substances. There is however still an inadequate understanding of the mechanisms of nanocarrier improvement of brain effects in vivo (Vendittoand and Szoka, 2013; Petrosand and DeSimone, 2010).

Distribution into the brain is carefully regulated by the blood–brain barrier (BBB). The endothelial cells enclosing the brain capillaries establish both a structural and a functional barrier with tight junctions, increased expression of efflux transporters, and reduced vesicular transport (Abbott, 2013). The brain can be reached only through very restricted paracellular transport, passive diffusion or by saturable transport, such as with transporter proteins, receptor mediated transcytosis or adsorptive endocytosis (Abbott, 2013). Nanocarriers can be designed to specifically target these processes, but little is known about the actual BBB passage in quantitative terms (Lajoieand and Shusta, 2015; Lindqvist et al., 2013).

The concentration–time profile in the brain of a drug administered in a nanocarrier will be determined both by the transport mechanism of the carrier at the BBB and by the drug itself. The rate and extent of drug exposure in the brain are generally determined by many factors including protein binding, binding to brain tissue, passive membrane permeability, active uptake and efflux transport, interstitial fluid (ISF) bulk flow and metabolism in the endothelial cells or brain (Hammarlund-Udenaes et al., 2008). When the drug is encapsulated into liposomes or bound to a nanoparticle, factors such as distribution, active and passive transport processes acting on the nanocarrier and release rate in the different sites, will also affect the pharmacological response in the brain. Some of these nanocarrier related aspects can be optimized in the lab depending on the desired outcome, e.g. the nanocarrier composition will determine the release rate (Ait-Oudhia et al., 2014). However, in order to give a pharmacological effect, the substance has to be available at the target receptor in its active form, i.e. either released from the nanocarrier at a sufficiently high rate to result in pharmacologically relevant concentrations, or with the binding domain of the drug available for the receptor, also this at relevant concentrations.

Nanocarriers are often evaluated based on percentage of dose that can be found in the brain tissue (van Rooy et al., 2011). This measure does not provide any information of the actual pharmacological active concentration at the receptors or the relative exposure of active drug in the systemic circulation that potentially could cause side effects.

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http://dx.doi.org/10.1016/j.ejps.2016.07.003
0928-0987/

The ratio of released, unbound drug in brain ISF compared to released, unbound drug in plasma, $K_{p,uu}$, can be a more valuable parameter to assess the nanocarriers, especially for drugs with target receptors that are extracellularly located in the brain. This however requires measuring the unbound drug in plasma and brain separated from the drug present in the liposomes/nanoparticles, which can be made using microdialysis (Lindqvist et al., 2013; Lindqvist et al., 2015).

To understand the net effect of the different processes on the concentration–time profile of active drug in the brain, the factors controlling the brain distribution of both the carriers and the free substance can be combined and evaluated using a model based approach. Modeling and simulation is a powerful tool to understand BBB transport and brain distribution of drugs (Syvanen et al., 2006; Goldenand and Pollack, 1998; Sjostedt et al., 2014). This has great potential in the nanocarrier field. There are some examples of physiologically based modeling and more empiric approaches concerning nanocarrier formulations in the literature (Ait-Oudhia et al., 2014; Li et al., 2010). Unfortunately the conclusions that can be drawn are limited, due to lack of quantitative data, e.g. separation of released and encapsulated drug.

The purpose of this study was to identify key factors that determine the effective drug concentration–time profile in brain, when using a nanocarrier formulation, as exemplified with a liposomal preparation. Nanodelivery with liposomes is a safe mode of administration that has been approved for human use (Barenholz, 2012; Gaillard et al., 2014). The paper includes investigation of which properties of the carrier that are of importance for increasing brain concentrations. We also wanted to highlight which kind of small molecular compounds could benefit the most from a nanocarrier formulation. To address these questions, we present a mechanism-based model describing systemic pharmacokinetics, release and BBB transport of a drug in a liposomal formulation. The model is based on data from a preclinical study in rats, showing increased brain delivery with the liposomal formulation compared to when free drug is administered (Lindqvist et al., 2013). Data included total liposomal concentrations in plasma as well as released, free drug in blood and in brain ISF. Alternative models for describing the increased brain distribution are explored. In addition, simulations of varying compound and carrier characteristics were performed. The model can serve as a tool for understanding uptake, release and disposition of liposomal preparations and support decision making when designing new carrier–drug combinations.

2. Methods

2.1. Source of experimental data

Preclinical data on the opioid peptide DAMGO encapsulated into PEGylated liposomes was used as a basis for the model building (Lindqvist et al., 2013). In this dataset, total drug in plasma, released drug in blood, as well as the released drug in brain ISF were available. The liposomes were composed by egg-yolk phosphatidylcholine (EYPC), and cholesterol (4:3) and were coated with GSH-PEG-DSPE (glutathione 1,2-distearoyl-sn-glycero-3-phosphoethanolamine conjugated polyethylene glycol, MW 2000). The average size of the liposomes was 127 nm and the polydispersity index was 0.024. Average data from 12 rats receiving 12.5 mg/kg of the DAMGO liposome formulation as a 10 minute infusion was included. Plasma was sampled at 0, 5, 9, 110, 290 and 470 min after the start of infusion and provided information of the total concentrations of DAMGO (encapsulated plus free) in plasma. Free blood concentration from the jugular vein and ISF (striatum) of the brain were sampled in 20 minute fractions with microdialysis throughout the experiment.

2.2. Model for systemic disposition

The model describing the systemic disposition (plasma concentration time profile) of the liposomal formulation was based on a two compartment model with one compartment for the encapsulated drug in plasma and one compartment for the free, released drug in plasma. This part of the model was kept as simple as possible to focus on the BBB transport. Release from the liposomal compartment to the corresponding free drug compartment was described with the release rate constant k_{rel}. The volumes of distribution of the unbound drug in the compartments were described by $V_{c,free}$ and $V_{c,lipo}$, respectively, and elimination of liposomes themselves including their drug content was described by CL_{lipo}. Different models to capture the initial peak of free DAMGO in plasma were compared visually. This included models describing different features of the formulation (first order release from the liposomes, a part of the dose as free drug already from the start, and a fast release fraction from the liposomes), as well as disposition with the addition of a peripheral liposomal compartment (e.g. that the liposome itself distributes into tissues).

2.3. Model describing the brain distribution

The brain distribution was described with the addition of an endothelial cell compartment and an ISF brain compartment added to the systemic model. The passage of free drug across the endothelial cell membranes was described with passive diffusion and active transport. The passive clearance ($CL_{passive}$) was assumed to be the same in both directions and across both the luminal and abluminal membranes. Active transport clearance was included in the model in the form of efflux from the endothelial cytosol to blood (CL_{efflux}). One compartment was used to describe the intra brain distribution, assuming that intracellular distribution and binding to brain tissue is not the rate limiting step. Brain ISF bulk flow was described with CL_{bulk}. This elimination pathway was assumed not to influence the central plasma compartment, since the amount of drug that is cleared this way will be negligible compared to the amount in plasma. No metabolism in brain or endothelial cells was included in the model. The volumes of distribution of the unbound drug in the endothelial cell and ISF brain compartments were described by V_{ec} and $V_{u,brain}$, respectively.

Various models for describing the mechanism of the increased brain uptake that was observed experimentally for the liposomal formulation were evaluated. Possible transcytosis of the liposome across both the luminal and abluminal membranes into a liposomal brain compartment was described by $CL_{transcytosis}$ (Model a). Endocytosis of the liposome across the luminal endothelial membrane into a liposomal endothelial compartment was described by an endocytosis clearance, $CL_{endocytos}$ (Model b). The release rate constant, k_{rel}, describing the release from the liposomal compartment to the free drug was assumed to be the same as in the central compartment to keep the model as simple as possible. An alternative mechanism with transfer from the liposomal central compartment directly into the free endothelial cell compartment was described by a "fusion clearance" (CL_{fusion}) which could illustrate the release of drug into the endothelial cell when liposomes merge with the phospholipids in the cell membrane (Model c). The possibility of increased brain permeability and inhibition of active efflux caused by the liposomal formulation was described/illustrated by alterations in $CL_{passive}$ (Model d) and CL_{efflux} (Model e), respectively. The transcytosis, endocytosis and the fusion clearance models (a–c) essentially all describe similar processes of liposomes being delivered into the endothelial cell, but the path length that the liposome stays intact varies. The increased passive permeability model (d) and the efflux inhibition model (e) both illustrate liposomal influence on the BBB function.

2.4. Parameterization and settings for modeling of liposomal DAMGO

The compound specific parameters for DAMGO were set to previously published values (Lindqvist et al., 2016). The net influx clearance from blood to brain (CL_{in}) of 1.05 μL/min/g_brain and the net efflux clearance (CL_{out}) of 14.1 μL/min/g_brain were used to calculate the