Powder Technology 284 (2015) 1–11

Contents lists available at ScienceDirect

Powder Technology

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

Optimization of spray drying process for formulation of solid dispersion containing polypeptide-k powder through quality by design approach

Puneet Kaur, Sachin Kumar Singh *, Varun Garg, Monica Gulati, Yogyata Vaidya

School of Pharmaceutical Sciences, Lovely Professional University, Phagwara 144411, Punjab, India

ARTICLE INFO

Article history:
Received 22 January 2015
Received in revised form 10 June 2015
Accepted 13 June 2015
Available online 20 June 2015

Keywords:
Polypeptide-k
Poor solubility
Spray drying
Solid dispersion
Box Behnken design
ANOVA

ABSTRACT

Polypeptide-k is an antidiabetic phytochemical isolated from dried and ripened seeds of *Momordica charantia*. The peptide has not been able to find much clinical use despite its established therapeutic effect. This is mainly attributed to its poor aqueous solubility. Present study presents the formulation of solid dispersion containing polypeptide-k to enhance its aqueous solubility. The study was carried out by employing the optimization approach using spray drying technique. Variables were evaluated using Box Behnken design. These include trehalose to drug ratio, tween-80 to drug ratio, inlet air temperature and feed flow rate. Responses measured were moisture content, solubility, product yield and angle of repose. Data analysis by ANOVA test indicated that ratio of trehalose to PPK significantly affected product flow properties and yield, whereas ratio of tween-80 to PPK was found to significantly affect moisture content, solubility and flow properties of the resultant formulation. Inlet air temperature significantly influenced moisture content while feed flow significantly affected moisture content and product yield. The optimized batch of formulation exhibited higher solubility in water as well as various aqueous buffers as compared to pure polypeptide-k. Through PXRD and SEM, it was inferred that enhanced solubility was due to reduced particle size and increased surface area of polypeptide-k present in the formulation.

1. Introduction

Polypeptide-k (PPK) is obtained from the dried seeds and ripened fruits of *Momordica charantia*, family Cucurbitaceae [1]. This phytochemical is known for its anti-diabetic potential and has gained importance in therapy of diabetes in last few decades. It is available in Asian markets as sublingual tablets [1,2]. The past decade has witnessed extensive research in order to fully explore its antidiabetic potential. PPK shows high homology to human insulin and has been reported to activate the inactive insulin [1]. In addition, it is also known to rejuvenate pancreas [1]. Till date, the goal of oral delivery of PPK has not been achieved. Major impediment in this direction has been poor aqueous solubility of PPK which results in its reduced bioavailability. Several approaches have been explored to enhance solubility and dissolution rate of poorly soluble drugs. These include, particle size reduction to micron and submicron levels [3,4]; complexation with cyclodextrins [5]; preparation of metastable polymorphs [6]; nanoemulsion; conversion of crystalline drug candidates to amorphous one [7–9]; liquisolid compacts [10]; and solid dispersion [11]. However, all techniques suffer from one or more limitations. Micro or nanoparticles tend to undergo Ostwald's ripening during storage, which leads to increase in particle size. Thus in order to improve the stability of micro or nanoparticles concentration of steric stabilizers like hydroxypropylmethyl cellulose (HPMC), hydroxypropyl cellulose (HPC), povidone (PVP K-30), pluronics (F68 and F127) and electrostatic stabilizers like surfactants (polysorbate (tween-80), sodium lauryl sulfate (SLS)), are required to be added to the formulation [12]. These additives adversely affect the safety profiles of these formulations [4,13]. Amorphous systems are thermodynamically unstable and tend to recrystallize during manufacturing or storage [14,15].

Cyclodextrins have generated interest in the fields of pharmaceuticals due to their ability to modify physical, chemical and biological properties of hydrophobic drugs. Cyclodextrins form inclusion complexes with these drugs [16,17]. However, due to their high molecular weight, relatively low water solubility and possible parenteral toxicity, the amount of cyclodextrins that can be used in most pharmaceutical formulations is limited [18].

Solid-dispersion technique is a simple and economical method and has, till date, proven to be the most successful in improving the dissolution and bioavailability of poorly soluble, active pharmaceutical ingredients. Preparation of solid dispersion involves deposition of drug on the surface of an inert carrier resulting in larger surface area of the drug. This leads to a faster dissolution rate. A variety of hydrophilic inert substances with high surface area have been employed for the preparation of solid dispersions [11,19]. Commonly reported excipients used in preparation of solid dispersion include propylene glycol (PG), polyethylene glycol (PEG) 4000, 6000 and 8000, tween-80 and povidone (PVPK-30) [11]. For converting solid dispersed powders into a free flowing powder and protecting them from heat generated during drying process, suitable

* Corresponding author: Tel.: +91 9888720835; fax: +91 1824501900.
E-mail addresses: singhsachin23@gmail.com, sachin_pharma06@yahoo.co.in (S.K. Singh).

http://dx.doi.org/10.1016/j.powtec.2015.06.034
0032-5910/

Table 1
Variables for Box–Behnken study.

Independent factors		Design level	
Uncoded	Coded	Uncoded	Coded
Carrier to drug ratio (w/w)	A (X1)	0.050–1.0	−1
		0.075–1.0	0
		0.100–1.0	+1
Solubilizer to drug ratio (w/w)	B (X2)	0.10–1.0	−1
		0.15–1.0	0
		0.20–1.0	+1
Inlet air temperature (°C)	C (X3)	120	−1
		125	0
		130	+1
Feed flow rate (ml/min)	D (X4)	16	−1
		18	0
		20	+1

carriers like sugars are added into the formulation. Lactose, mannitol, sorbitol and trehalose etc. [20], have been commonly used for this purpose. They have been reported not only to improve flow properties of the powders but also to improve their stability (as cryoprotectant) as well as solubility [21,22]. The technique has proven to be very useful for protein and peptide drugs.

The techniques that are routinely used for the preparation of solid dispersions include solvent evaporation method, melt extrusion, kneading, co-grinding as well as co-precipitation method [11,23]. Solvent evaporation technique is the most preferred choice because thermal decomposition of drugs or carriers can be prevented due to the relatively low temperatures required for the evaporation of solvents [24]. Choice of drying technique employed to evaporate solvent plays a major role in the product performance. On the other hand, a well-designed drying protocol also helps in improving the interaction of poorly soluble drug with the inert carriers, which ultimately results in increased solubility and better flow property of the drug. The drying processes routinely used are: rotary evaporation, freeze drying and spray drying. Among these, spray and freeze drying processes have shown better performance in terms of product yield and ease of scale up. These are, in particular, more suitable for drying of proteins and peptides.

The technique of solid dispersion has never been explored to enhance the aqueous solubility of PPK [25]. In the present study, an attempt has been made to develop and optimize a solid dispersion system of poorly soluble polypeptide-k using different drug to carrier ratios and altering the conditions for spray drying process. Final product has low water activity and lesser density which results in easy storage and transportation. Spray drying technique involves atomization of a liquid product in hot gas current to obtain instantaneous powder [26–29]. Air is the most preferred choice as a drying gas, however, in some of the cases nitrogen gas has also been used [30]. Physicochemical properties of the final product mainly depend on inlet temperature, air flow rate, feed flow rate and type of carrier used. The formulation and processing parameters were optimized using response surface methodology (RSM). RSM uses a group of mathematical and statistical techniques to investigate the relationships between response and the independent variables [31–33]. Box Behnken design was used to investigate ratio of drug to carriers, feed flow and inlet temperature in order to optimize the best formulation. Best formulation was said to be the one which had better solubility, good powder flow, better product yield and least moisture content.

Table 2
Formula composition of PPK solid dispersion system.

Ingredients	Quantity/batch (g)
Polypeptide-k	5
Trehalose	0.25–0.5
Tween-80	0.50–1

2. Materials and methods

2.1. Materials

Polypeptide-k (PPK) was isolated from dried seeds of ripened fruits of *M. charantia* by the procedure reported by Khanna (2004). Polyethylene glycol 200, 400, 600, 4000 and 6000, propylene glycol (PG), tween 20, 60 and 80, span 20, 40, 60 and 80 and Pluronic F-68 were purchased from Central Drug House (CDH), New Delhi, India. Formic acid, trehalose, mannitol and sorbitol were purchased from Lobachemie, Mumbai, India. Spray dryer, SprayMate, Jay Instruments and systems, Navi Mumbai, India was used for drying the solid dispersion of PPK. An UV–visible double beam spectrophotometer, UV-1800, Shimadzu, Japan was used for quantitative estimation of the drug. Magnetic stirrer, REMI, India was used for mixing of solutions.

2.2. Isolation of polypeptide-K from M. charantia

Dried seeds of *M. charantia* were collected from the ripened fruits. As polypeptide-k is a storage protein, it is found in more quantity in the dried seeds. Seeds were ground to a fine powder. The pulverized seeds were treated for de-oiling with a mixture of water and acetone in the ratio of 3:1. The dispersion was kept for 3h and then filtered through muslin cloth. The residue left over the muslin cloth was dried and suspended in a mixture of water and acetone. The resultant powder was dried and dispersed in quantity sufficient water and pH was adjusted to 9.5 by using ammonium hydroxide. The supernatant was collected by centrifugation and its pH was adjusted to 3 by using diluted sulphuric acid. The flocculent precipitate so formed was collected and dried. The dried mass was powdered and grounded powder was washed with a mixture of water and acetone (3:1) to remove oil, salts and undesirable material till this gave a single spot on HPTLC and TLC [1].

2.3. Preliminary studies to select the suitable solubilizers

PEG 200, 400, 600, 4000 and 6000, propylene glycol (PG), tween 20, 40, 60 and 80, span 20, 40, 60 and 80 and Pluronic F-68 were evaluated for their solubility enhancing effect on PPK. Liquid solubilizers were used as such, whereas, solid agents (1 g each) were dissolved in 10 ml water. In each case, 50 mg of PPK samples were added to 2 ml of solubilizer and the mixtures were vortexed for 2 min at regular time intervals for 48 h. Solutions were centrifuged at 3500 rpm for 15 min and the supernatant was collected. Suitable dilutions were made and solution was passed through 0.45 μm membrane filter. Filtered solutions were analyzed through a UV–visible double beam spectrophotometer at 272.23 nm.

2.4. Preliminary studies to select the suitable solid carrier

A suitable carrier is required to adsorb the liquid solution of drug properly and provide free flow without aggregation of the powder [20–22]. Trehalose, mannitol and sorbitol were evaluated for their flow improving properties. It was found from the solubility studies carried out in different aqueous and organic solvents that 10% v/v formic acid (in water) was the only solvent in which PPK was found to exhibit significant solubility. Though the percentage of formic acid used in the present study was just 10% v/v in water, which is quite low, for safety purpose mask and gloves were used to cover our body parts.

PPK (10 g) was added in 10% v/v formic acid solution containing 0.2% of tween-80. Solution was divided into three equal parts and trehalose, mannitol and sorbitol were added separately to one of the parts. Solutions were spray dried at an inlet temperature of 125 °C, feed rate 18 ml/min and aspiration air flow rate of 1200 rpm. Dried powder