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Research Paper

Combining inkjet printing and amorphous nanonization to prepare personalized dosage forms of poorly-soluble drugs

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ABSTRACT

Inkjet printing of drug nanosuspension on edible porous substrates was carried out for the first time with the objective of preparing personalized dosage forms of poorly soluble drugs. Amorphous drug–polysaccharide nanoparticle complex (or drug nanoplex in short) was used as the nanosuspension ink, instead of the conventional crystalline nanodrug. The amorphous drug nanoplex exhibited low propensity to Ostwald ripening growth, high colloidal stability, and supersaturation generation capability making it ideal for printing. Nanoplexes of ciprofloxacin – a BCS Class IV compound – prepared by complexation with dextran sulfate were used as the nanosuspension ink at two different sizes (i.e. ≈265 nm and 188 nm). Inkjet printing was performed on cellulose substrate at 0.25% (w/v) nanosuspension concentration and 5% (w/v) polyethylene glycol.

For both nanoplex sizes, the results indicated that the printed dose could be increased by increasing the number of droplets dispensed. However, exact correlations between the achievable dose and the number of droplets dispensed were not evident, which was likely caused by the spatial non-homogeneity in the nanosuspension concentration. Compared to the larger nanoplex, printed nanodrugs of the smaller nanoplex consistently exhibited higher payload with better batch-to-batch reproducibility (<6%). The maximum achievable payload was equal to ≈2.5 μg/cm^2, which was multifold higher than that achieved had inkjet printing of ciprofloxacin solution been performed. Nevertheless, print substrate with higher liquid uptake capacity is needed to increase the payload nearer to the therapeutic dose. Lastly, the drug release and non-cytotoxicity of the printed nanodrug were successfully established *in vitro*.

1. Introduction

Conventional oral solid dosage forms (i.e. tablets) are manufactured in limited discrete doses that require the patients to split the tablets when a smaller or fractional dose is required. The practice of splitting tablets leads to inaccurate dosing that causes adverse effects or inadequate therapeutic levels, particularly for tablets with enteric or controlled release coatings [1,2]. Personalized dosage forms enable the administration of non-discrete doses that are tailored to the individual patients depending on their gender, bodyweight, and age group [3]. Personalized dosage forms are particularly needed for pediatric patients because of the rapid changes in the physiological and metabolic functions in children, as well as for geriatric patients because of the existence of co-morbidity and changes in the gastrointestinal tract and renal clearance in the elderly [4].

Inkjet printing of drug solution has emerged as the preferred method to prepare personalized dosage forms by virtue of the mature state of the inkjet printing technology that enables printing of drug solution down to the picoliter quantities at high precision. Inkjet printing also facilitates on-demand preparation of personalized dosage forms at the points of care (e.g. hospitals, pharmacies), without the need for bulky and costly equipment [5]. Moreover, as

Abbreviations: BCS, Biopharmaceutics Classification System; CIP, ciprofloxacin; CV, coefficient of variation; DMEM, Dulbecco's modified Eagle's medium; DMSO, dimethyl sulfoxide; EDTA, trypsin–ethylenediaminetetraacetic acid; FESEM, field emission scanning electron microscopy; FTIR, Fourier transform infrared spectroscopy; GRAS, generally recognized as safe; HPLC, High Performance Liquid Chromatography; HPMC, hydroxypropyl methylcellulose; MTT, (3-(4,5-dimethyl thiazol-2-yl)-2,5-diphenyltetrazolium bromide); OD_{550}, optical density at 550 nm; PBS, phosphate buffer saline; PCS, photon correlation spectroscopy; PEG, polyethylene glycol; PXRD, Powder X-ray Diffraction; USP, United States Pharmacopeia; UV–Vis, ultraviolet–visible.

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multiple drugs can be printed onto the same substrates at different doses, inkjet printed drug represents an ideal solid dosage form to address polypharmacy in geriatric healthcare [3].

Numerous studies on inkjet printed drug have been carried out, where the initial studies focused on (i) demonstrating the technical feasibility of inkjet printed drug and (ii) physical characterizations of the printed drug products [6–9]. Subsequently, the effect of using different substrates on the batch-to-batch variations of the printed dose was investigated from which porous substrates were found to yield the best performance, particularly when multi-pass printing on the same substrate was needed to achieve the therapeutic dose [10,11]. Very recently, inkjet printing of drug solution was also employed to prepare thin drug films for taste masking purposes [12] and to print multiple drugs on transdermal microneedles [13].

The maximum achievable dose of the inkjet printed drug, however, is dictated by the drug solubility in the ink medium. Consequently, inkjet printing of poorly-soluble drugs, which represent a significant fraction of the newly discovered drugs and drugs currently in the developmental pipeline [14], remains impracticable due to the resultant printed dose that is far below the therapeutic dose. While organic solvents can be meticulously selected as the ink medium to increase the printed doses [15,16], it is often not feasible as many of these drugs are in fact poorly soluble in both water and most organic solvents [14].

For such drugs, the use of drug nanoparticle suspension as the ink medium, in place of the drug solution, was proposed by Pardeike et al. [17]. The use of nanosuspension ink was motivated by (i) the small size of nanoparticles that enabled them to pass through the inkjet nozzle and (ii) their large specific surface areas that enhanced the dissolution of poorly soluble drugs. The proof-of-concept study of Pardeike et al. [17], however, was limited to the development of the nanosuspension ink (using folic acid as the model drug) and generation of stable nanosuspension droplets from a piezoelectric dispenser, where printing on substrates was not carried out. As a result, the feasibility of employing nanosuspension ink to prepare personalized dosage forms of poorly-soluble drugs has not been properly assessed to date in terms of the printed dosage forms produced.

Not unlike inkjet printing of drug solution, the key to successful inkjet printing of drug nanosuspension lies in the nanosuspension ink itself, where nanosuspension exhibiting high and prolonged colloidal stability is required. In this regard, drug nanosuspension is typically prepared in its crystalline form by size reduction or antisolvent precipitation techniques [18]. Drug nanocrystals in their aqueous suspension form, however, are prone to solution-mediated growth due to Ostwald ripening [19]. As a result, drug nanocrystals are usually transformed to dry powders immediately after their preparation to prevent the Ostwald ripening growth. If the crystal grows in the nanosuspension ink, the increased nanoparticle size will inevitably cause nozzle blockage in inkjet printing. Therefore, nanocrystalline drug represents a less-than-ideal option for the nanosuspension ink.

The objective of the present work was to demonstrate for the first time inkjet printing of nanodrug onto edible porous substrates, using aqueous suspension of amorphous drug nanoparticles as the nanosuspension ink. The amorphous nanodrug was prepared by drug–polysaccharide electrostatic complexation technique developed previously in our group in the form of drug–polysaccharide nanoparticle complex (or drug nanoplex in short) [20]. Unlike drug nanocrystals, the amorphous drug nanoplex is highly stable in water, where it only dissolves in the presence of salt, for example in phosphate buffer saline (PBS), owing to the charge screening effect of the salt. As a result, the drug nanoplex is not prone to the Ostwald ripening growth making it suitable to be used as nanosuspension ink.

In addition, the other benefit of using the amorphous drug nanoplex lies in the potential bioavailability enhancement of the poorly-soluble drugs. The enhanced bioavailability is attributed to the supersaturation generation afforded by the amorphous form, resulting in apparent solubility that is multifold higher than that generated by the nanocrystalline form [21]. As a result, lower doses are needed to achieve the therapeutic effect, which translates to lower printed doses required.

In the present work, ciprofloxacin – a Biopharmaceutics Classification System (BCS) Class IV compound [22] – was used as the model drug that exhibits poor solubility at 25 °C in water (75 μg/mL) as well as in common pharmaceutical solvents (e.g. 60 μg/mL in ethanol, 70 μg/mL in isopropanol, and 140 μg/mL in acetone) [23]. Ciprofloxacin nanoplexes of two different sizes were prepared by complexation with dextran sulfate. The nanoplex was printed onto edible porous substrates made of hydroxypropyl methylcellulose (HPMC). The correlation between the achievable dose and the number of droplet dispensed was examined. The printed nanodrug was characterized in terms of the payload and its batch-to-batch variations, *in vitro* release profile, and cytotoxicity.

2. Materials and methods

2.1. Materials

2.1.1. Materials for inkjet printing

Ciprofloxacin (CIP), HPMC, high molecular weight polyethylene glycol (PEG 8000), phosphate buffer saline (PBS, pH 7.4), Pluronic F68, acetonitrile, and glacial acetic acid were purchased from Sigma Aldrich (USA). Dextran sulfate (MW 5 kDa) was purchased from Wako Pure Chemical (Japan).

2.1.2. Materials for cytotoxicity test

Dulbecco's modified Eagle's medium (DMEM) and fetal bovine serum were purchased from HyClone (Thermo Scientific, USA). 0.25% trypsin–ethylenediaminetetraacetic acid (EDTA) solution, p enicillin–streptomycin, dimethyl sulfoxide (DMSO), and (3-(4,5-di methylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) (MTT, 98% purity) were purchased from Gibco (Canada), PAA Laboratories (Austria), Sigma–Aldrich (USA), and Alfa Aesar (UK), respectively. A549 adenocarcinomic human alveolar basal epithelial cells were purchased from ATCC (USA).

2.2. Methods

2.2.1. Preparation and characterizations of amorphous drug nanoplex

In the drug–polysaccharide complexation technique, ionized drug molecules were mixed with oppositely charged polysaccharides to form soluble drug–polysaccharide complex. Owing to the hydrophobic interactions between the drug molecules, aggregates of the drug–polysaccharide complex were formed and subsequently precipitated out to form the drug nanoplex upon reaching a critical concentration, which was governed by the drug hydrophobicity. Amorphous nanoparticles were formed because the strong electrostatic interactions between the drug and polysaccharides inhibited the former from assembling into ordered crystalline structures.

CIP was dissolved at 1.0% (w/v) in 0.2% (v/v) aqueous acetic acid solution and dextran sulfate was dissolved in deionized water at 0.45% (w/v) together with 0.2% (w/v) Pluronic F68. The CIP solution was then added to the dextran sulfate solution under gentle stirring at ambient condition. The resultant solution was let sit for 30 min for the complexation to equilibrate. Afterward, the CIP nanoplex was recovered by three cycles of centrifugation at 13,000×*g* and 5 min and re-suspended in deionized water to be