Electrospun Nanofiber Matrix with a Mucoadhesive Backing Film for Oramucosal Drug Delivery

Charu Tyagi, Lomas Tomar, Yahya E. Choonara, Lisa C. Du Toit, Pradeep Kumar, and Viness Pillay

***Abstract*—A rapidly disintegrating oramucosal drug delivery system was prepared with mucoadhesive polymeric backing film. Various polymers were investigated for suitability in the development of electrospun nanofibers layer and different set of polymers for backing film layer. Individual layers and the system with nanofiber spun directly on the backing film were characterized using standard techniques. Ex vivo drug permeation analysis was performed to establish the suitability of the system. Polyvinylalcohol (PVA) produced drug-loaded fibers with the most acceptable morphology and a disintegration time of 5sec. Acceptable films casted were of PVA/hydroxypropylmethylcellulose (HPMC), with required mucoadhesive property and average disintegration time of 7-60sec. Diphenhydramine loaded delivery system exhibited 42-82% drug permeation in study time period potentiating the feasible application of the system for oramucosal drug delivery.**

***Index Terms*—Backing film, electrospun nanofiber matrix, mucoadhesion, oramucosa drug delivery.**

I. Introduction

A drug delivery system intended for accelerated oramucosal drug delivery is required to be rapidly disintegrating, releasing drug almost instantaneously to the buccal mucosa for immediate absorption. This may be achieved by the use of water-soluble polymers and a large surface area exposed to the dissolution medium [1], [2]. Electrospun fibers exhibit an exceedingly large surface area to mass ratio, which not only enhances the dissolution rate, but also increases the bioavailability and total amount of drug released released and may therefore find application in rapid oramucosal drug delivery.

Due to the relatively short residence time of an oramucosally administered drug delivery system at the site of absorption, mucoadhesion is often required [3]. Mucoadhesive drug delivery systems are advantageous in that the entire system is rendered immobile, an intimate contact between the system and buccal mucosa is created and a high drug concentration at the absorption surface is achieved [3]. This results in a reduction in the required drug concentration as well as an improved bioavailability [4]. Thin, mucoadhesive films are favourable for oramucosal drug delivery due to the flexible nature and high contact surface area of such films [3]-[5].

The paper presents development of a mucoadhesive polymeric backing film layer and a drug-loaded electrospun fiber layer with the aim of producing a flexible, mucoadhesive fibrous matrix system by depositing the latter directly onto the former. The film layer provided a flexible, mucoadhesive backing layer with a large exposed surface area and the drug-loaded fiber layer exhibited an exceedingly high surface area, due to the fiber dimensions, allowing for more rapid disintegration and drug release than what could be achieved from a drug-loaded film formulation. Polymers investigated for the development of the backing layer were selected based on water-solubility and film or membrane-forming propensity. The polymers investigated for the fibrous layer were selected based on water-solubility, mucoadhesiveness and electrospinnability. Diphenhydramine (DPH) was chosen as the model drug to assess the loading and release efficacy of the delivery system.

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II. Materials

Polyvinylalcohol (PVA) (87-89% hydrolyzed, Mw 13,000-23,000g/mol), poly (ethylene oxide) (PEO), poly (acrylic acid) (PAA) (Mw 1,800g/mol) and diphenhydramine (DPH) were purchased from Sigma-Aldrich (St. Louis, Missouri, USA). Propan-2-ol, glycerol, citric acid, sodium carbonate, sodium chloride and ethanol were purchased from Rochelle Chemicals (Johannesburg, South Africa). Hydroxypropylcellulose (HPC) (Klucel Type EF and Type HF) was purchased from Hercules (Wilmington, Delaware, USA). Hydroxypropylmethylcellulose (HPMC) was purchased from Colorcon Limited (London, England).

III. Experimental

A. Casting Polymeric Backing Film

Two films were casted and investigated as backing film: PMC and the PVA/HPMC films. Polymer solutions were prepared by dissolving HPMC (0.5-3%w/v) or the PVA (0.5-2.5%w/v) and HPMC (0-1%w/v) in a 2:1 and 4:1 mixture of deionized water and propan-2-ol respectively. Glycerol was added to both the solutions as a plasticizer at varying concentrations of 5-70% of the polymer mass. Solutions were syringed into the respective rectangular, flat-bottomed molds and placed under an extractor at 21 ℃ for 48 hours for complete solvent evaporation and film formation to occur.

B. Electrospinning Nanofiber Matrix

Solutions, employing different solvents and polymers at varying concentrations, were prepared in order to assess the electrospinning. Once appropriate solvent/polymer

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combinations were determined, diphenhydramine (DPH) was dissolved in the polymer solution. Solutions were placed in a 5mL pipette, which was fitted into the adjustable supporting bracket of an electrospinning device. Electrospinning of the solutions was using a custom-built electrospinning device (RGC Engineering, Johannesburg, South Africa). Fibers were collected on aluminum foil-lined board or on prepared polymeric backing films secured to the board. Polymers investigated for electrospinning were PVA, HPC and PEO. The solution preparation and electrospinning parameters are detailed in Table I. Glycerol or glycerin was used in all electrospinning solution as the plasticizer and citric acid as a taste-masking agent in PVA solution.

TABLE I: ELECTROSPINNING POLYMERIC SOLUTIONS AND PARAMETERS

Polymer	Solvent	Solution concentrations (%w/v)	Tip to collector distance (cm)	Applied voltage (kV)
PVA	W/P 2:1	15-30	5-11	15-20
HPC	W/P 2:1	10	5-8	20
PEO	Water	10	7-10	18-20

W/P: Deionized water/propan-2-ol

C. Characterization

The surface morphology of the electrospun fiber layer was analyzed by scanning electron microscopy (SEM), using a Phenom Microscope (FEI Company, Hillsboro, Oregon, USA). This was also used to confirm nanofiber formation. Rheological properties of PVA solutions employed in electrospinning were determined at 25˚C with the use of a Haake Modular Advanced Rheometer System (ThermoFisher Scientific, Karlsruhe, Germany). The shear rate was ramped from 0 to 500/s and viscosity and shear force were quantified for each solution. Mucoadhesion testing was performed on backing film using a *TA.XTplus* Texture Analyser (Stable Micro Systems, England) fitted with a cylindrical probe. Porcine buccal mucosal tissue was attached to the probe, using rubber bands, and exposed to simulated saliva (pH 6.75). The samples were attached to the stage directly below the probe. Mucoadhesion was tested by measuring the work of adhesion (WA) between the buccal mucosa and the film. The pre-test, test and post-test speeds were 2, 2 and 10mm/s, respectively. An applied force of 50g, a trigger force of 5g and contact time of 5 seconds were used for the test.

D. Disintegration Time of the PVA Fiber Layer and Backing Film Layer

The *in vitro* disintegration times were determined according to a modified method based on the United States Pharmacopoeia (USP) method for tablet disintegration testing using a Type PTZ 1 basket-rack assembly disintegration apparatus (Pharma Test, Hainburg, Germany). The disintegration medium was 150mL simulated saliva (pH 6.75) in a glass jar placed in a water bath maintained at 37˚C. Samples were cut into sections and placed on the mesh of the basket rack assembly, with a mesh disc placed on top. The basket rack assembly was raised and lowered through a distance of 55mm and at a frequency of 25 cycles per minute.

E. Drug Entrapment and Ex Vivo Drug Permeation Studies

Samples of the drug-loaded fiber layer were cut into sections, dissolved in simulated saliva (pH 6.75) and the drug (DPH) content of each section was analyzed by UV spectrophotometry (Specord 40, Analytik Jena, AG, Germany), at wavelength of 254nm.

PVA fiber loaded with DPH, were electrospun directly onto films made up of either PVA or PVA/HPMC. The resulting fiber-on-film systems were cut into sections. Sections of freshly excised porcine mucosa tissue were mounted in Franz Type Diffusion Cells (Perme Gear, Inc., Hellertown, Pennsylvania, USA) and equilibrated for 0.5h at 37˚C by adding PBS (pH 7.4) to both the acceptor and donor compartments. After equilibration, the PBS in the donor compartment was removed and replaced with a drug-loaded sample in simulated saliva (pH 6.75). Samples drawn from the acceptor compartment at different time intervals were analyzed by UV spectrophotometry, and replaced by the same volume of fresh PBS.

IV. RESULTS AND DISCUSSION

A. Polymeric Backing Films

HPMC films displayed a desirable thickness and uniformity at HPMC concentrations of 1-2%w/v. The ideal glycerol concentration for flexible film formation was 15-30%w/w of the polymer mass. These films easily torned, which made removal from molds problematic. PVA/HPMC films were thin and clear or cloudy, depending on the solution constituents. PVA concentrations between 0.5 and 2%w/v produced films of a desirable thickness at a fill volume of 50mL. Above this concentration the films were too thick and fiber deposition was not uniform on these films. The optimal glycerol concentration ranged between 10-15%w/w, below which the films were fragile and above it the films were difficult to work with. HPMC concentrations of 0-0.5%w/v worked well without causing breakage on removal of films from molds. The acceptable fill volume was found to be between 40 and 100mL. Below 40mL, film formation was not particularly even.

B. Electrospun Nanofiber Matrix

Electrospinning PEO solutions resulted in a thick, white layer on the collector surface. Nevertheless, fiber deposition was erratic and inconsistent having patches with great variation in thickness and even displaying clear regions. HPC electrospinning formed a fairly thick white layer on the collector, suggesting adequate fiber production. However, addition of drug hindered both fiber production and formation considerably. Therefore, no further studies were conducted on PEO and HPC for electrospinning. PVA electrospinning solutions exhibited optimum viscosity and most desirable fiber formation. The produced fibers formed a thick layer on the foil or film onto which they were electrospun, which is considered adequate for drug-loading. High plasticizer concentrations hindered the production of fibers, however lower concentrations did not interfered with

fiber production and rather resulted in a more flexible fiber layer. Drug incorporation had little or no effect on fiber formation. Electrospun PVA fibers were therefore deemed to be satisfactory for drug delivery. The ideal PVA concentration for the electrospinning solution was determined to be 25%w/v, the DPH concentration was 10%w/v and the excipients glycerol and citric acid at 5%v/v and 2%w/v, respectively.

C. Characterization

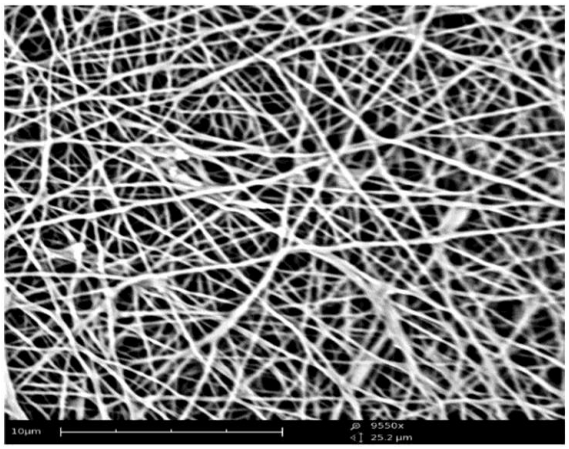

Fig. 1. Scanning electron micrograph of DPH loaded PVA fibers.

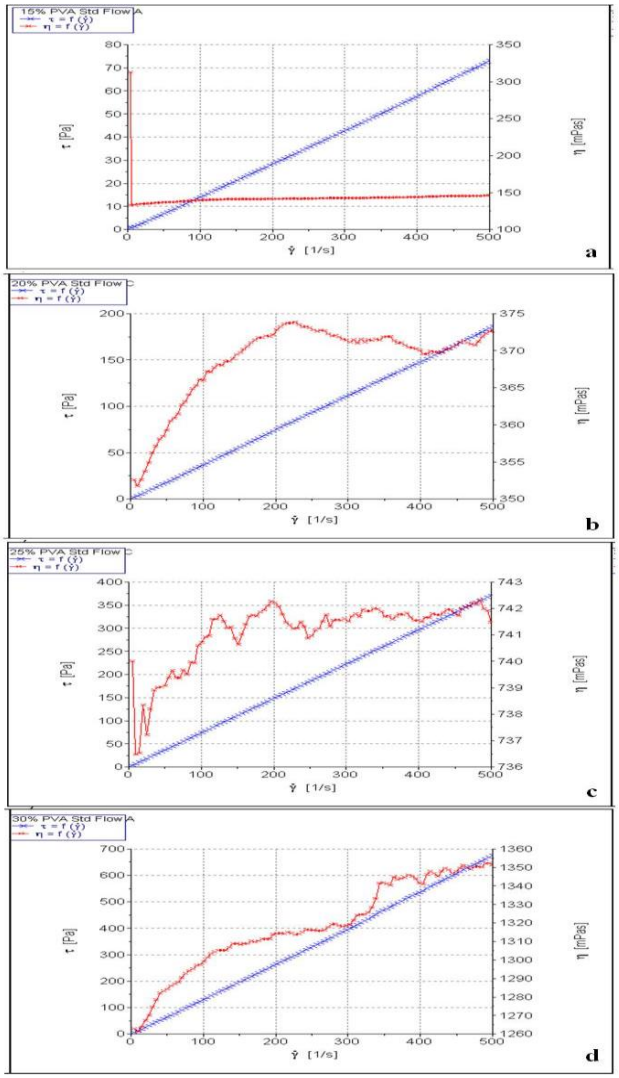

Fig. 2. Rheological profiles of (a) 15%w/v, (b) 20%w/v, (c) 25%w/v and (d) 30%w/v PVA solutions in 2:1 deionized water and propan-2-ol.

SEM analysis of PVA electrospun matrix clearly displays the presence of randomly oriented fibrous structures in the mesh (Fig. 1). Fibers formed were less than 500nm thick with somewhat uniform diameter and structure. Pores were also apparent between individual fibers.

The stress-srrain rheological parameters of a solution can have a substantial effect on the process of electrospinning as well as the quality and morphology of fibers that are formed. The degree of polymer chain entanglements, and hence the polymer concentration, has a considerable influence over the viscosity and electrospinnability of a solution. The actual conformation of individual polymer chains also has a significant influence on solution viscosity, considering that solutions containing coiled chains have a lower viscosity than those with extended chains [6]. It is therefore important to investigate the rheological properties of polymeric solutions employed in electrospinning. The rheological profiles of the 15, 20, 25 and 30%w/v PVA solutions in 2:1 deionized water and propan-2-ol are depicted in Fig. 2a-d.

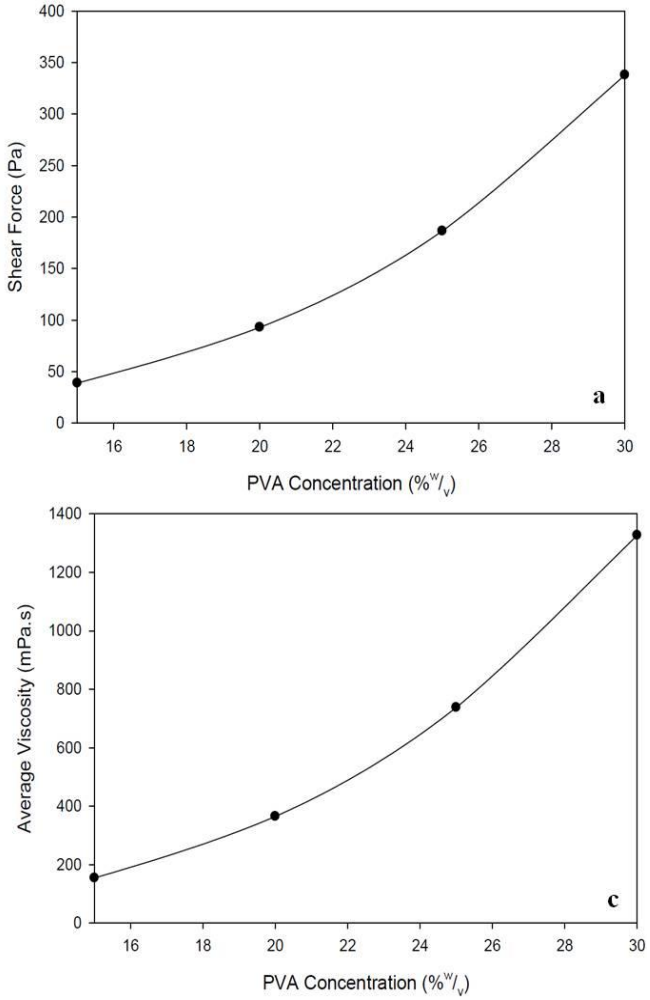

Fig. 3. Comparison for the electrospinning solutions with varying PVA concentrations (a) of average shear force; (b) of average viscosity.

The viscosity of the PVA solutions of different concentrations increased with increasing shear rate and appeared to exhibit non-Newtonian, dilatant flow properties. The average shear force (τ) and viscosity (η), over the shear rate range, plotted against PVA concentrations is presented in Fig. 3a-b. Figures appear to display a similar shaped curve, suggesting that the increase in average viscosity with

increasing polymer concentration is proportional to the increase in shear force. At PVA concentrations below 25%w/v, electrospun fibers did form properly and spraying of solution droplets occurred intermittently as the viscosity was too low for constant fiber jet formation. At PVA concentrations above 25%w/v, the solution was too viscous to pass through the capillary tip consistently. Fiber formation was satisfactory at a PVA concentration of 25%w/v.

Mucoadhesiveness is an eminent factor to consider in oramucosal drug delivery because the retention of a drug delivery system at a specific site influences drug absorption at that site [4]. The average work of adhesion (WA) was calculated from textural profiles. It was observed that as the HPMC concentration in the formulations increased above 0.25%, the average WA decreased. The process of mucoadhesion occurs largely through interpenetration and hydrogen bonding between the polymeric system and the mucosal surface. In order for interpenetration to occur between these two surfaces, the polymer chains are required to be flexible [4]. HPMC is able to form weak hydrogen bonds with mucosal surfaces and an adherent gel upon hydration, which both make it a strongly mucoadhesive polymer in theory. Notwithstanding this, it has a high glass transition temperature (>200˚C), which results in poor chain flexibility and therefore poor experimental mucoadhesion [7]. This elucidates the observed decrease in WA with an increase in HPMC concentration. However, formulations containing 0.5%w/v HPMC displayed an increase in WA with an increase in glycerol concentration. At the higher HPMC concentration, the increase in WA with increasing glycerol concentration may be attributed to the pliability of poorly-flexible HPMC chains being enhanced, hence resulting in an increased WA as the glycerol concentration was increased [8].

D. Disintegration Time Studies

The average disintegration time of the electrospun PVA fiber matrix was recorded as 5 seconds, which is desirable for a rapidly disintegrating drug delivery system. The average disintegration time for the backing film ranged between 7-60 seconds, depending on constituents, in the range used. To ensure high drug concentration at the absorption surface and prevent swallowing of the drug, the backing film layer is required to remain intact for a longer period of time than the fiber layer, holding fiber matrix at the absorption site.

E. Drug Entrapment and Ex Vivo Drug Permeation Studies

The average quantity of drug entrapped per 1.5cm^2 section of the electrospun fiber matrix varied according to PVA and drug concentrations in the electrospinning solutions and time of electrospinning. When these factors were increased, drug entrapment increased linearly. The average quantity of drug entrapped per section ranged between 0.3-7mg. Drug entrapment and fiber production was observed to be greater at low humidity. Researchers have reported that as the relative humidity increases, fiber diameter decreases and bead-formation sets in because solvent evaporation is retarded at higher humidity [9]. The technique of electrospinning is therefore sensitive to environmental conditions and, hence, must be conducted in a controlled environment. *Ex vivo* drug permeation study determines the expediency of employing a particular DDS for buccal administration. Fig. 4 depicts the drug permeation profiles of nanofiber matrix DDS. It was observed that 42-82% of the loaded DPH dose had passed through the buccal mucosal during the testing period of 90min.

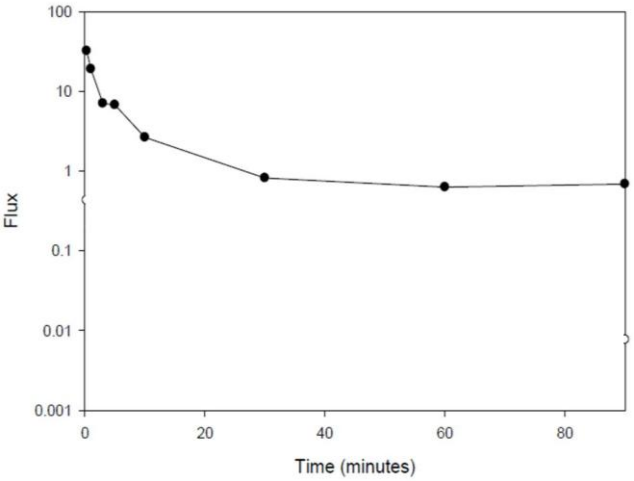

Fig. 4. Flux profiles of diphenhydramine (DPH). (in all cases SDs < 0.02, N = 3).

V. Conclusion

A polymeric film layer, containing PVA, HPMC and glycerol in concentrations of 1%w/v, 0-0.5%w/v and 10-15%w/w (of total polymer mass), respectively and a fill volume of 40-100mL were deemed acceptable for backing film production. Various polymers were investigated in order to develop a drug-loaded electrospun fiber layer for the system and PVA was identified as the most suitable polymer. DPH adopted as the model drug exhibited 42-82% permeation during the testing period. The ideal PVA concentration for the electrospinning solution was determined to be 25%w/v, the drug loading concentration was 10%w/v and the excipients citric acid and glycerol at 2%w/v and 5%v/v, respectively. This optimized electrospun nanofiber matrix system with polymer film backing proves ideal for drug delivery through oramucosa.

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