Development and Characterization of a Hydrogel Containing Silver Sulfadiazine for Antimicrobial Topical Applications

KARIN S. P. JODAR,[1] VICTOR M. BALCÃO,[1,2,3] MARCO V. CHAUD,[1] MATTHIEU TUBINO,[3] VALQUÍRIA M. H. YOSHIDA,[1] JOSÉ M. OLIVEIRA JR,[1] MARTA M. D. C. VILA[1,3]

[1]LaBNUS – Biomaterials and Nanotechnology Laboratory, i(bs)[2] – **I**ntelligent **B**io**s**ensing and **B**iomolecule **S**tabilization Research Group, University of Sorocaba, Sorocaba, São Paulo, Brazil

[2]CEB – Centre of Biological Engineering, University of Minho, Braga, Portugal

[3]Institute of Chemistry, University of Campinas, Campinas, São Paulo, Brazil

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ABSTRACT: Development and optimization of a hydrogel with impregnated silver sulfadiazine was pursued, for antimicrobial topical applications. The selected hydrogel exhibited a homogeneous appearance, with whitish colloration and devoid of any fractures or cracks. The content in impregnated silver sulfadiazine was within established limits (1%, w/w) with a standard deviation of up to 1.28%. The hydrogel presented a good characteristic in relation to release of the active antimicrobial principle, verified through swelling tests and antimicrobial activity. The swelling tests indicated a higher increase in weight during the first 6 h of contact with a moist environment, with a maximum value of 266.00 ± 0.81, and with maintenance of the original shape of the hydrogel. The impregnated silver sulfadiazine presented antimicrobial activity, as expected, indicating a prolonged release of the drug. The infrared spectra of the hydrogel with impregnated silver sulfadiazine indicated that the drug did not engage in any bonds with the polymeric matrix, which otherwise could have reduced its antimicrobial activity. The mechanical resistance tests produced good results, indicating that the hydrogels may be utilized in different locations of the human body with skin lesions. © 2015 Wiley Periodicals, Inc. and the American Pharmacists Association J Pharm Sci 104:2241–2254, 2015

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INTRODUCTION

Among the various dosage forms developed as modified-release systems, hydrogels can be considered controlled-release systems for drugs of topical application, depending on both the type of formulation and application in which they are used.[1] Hydrogels are three-dimensional (3D) networks of hydrophilic polymers that, in contact with water, swell and may release the drug by different mechanisms.[2] In this sense, hydrogels are of particular interest in the treatment of topical wounds because of their intrinsic low toxicity, potential for extended release of drugs, and the ability to keep the wound hydrated.[3–5] Review studies have pointed toward the use of various types of natural and/or synthetic polymers for the preparation of hydrogels, such as poly(acrylic acid), poly(ethylene oxide), poly(ethylene glycol) (PEG), poly(vinyl pyrrolidone), polyvinyl alcohol (PVA), polyglycerol esters of fatty acids, carbomers, sodium alginate, chondroitin sulfate, pectin, dextran, carboxymethylcellulose, and other cellulose derivatives, chitosan, gelatin, and gums.[6–11] PVA is extensively used to produce hydrogels aiming at the treatment of topical ulcers. PVA is a nontoxic, biocompatible polymer that has excellent film-forming properties and ease of processability, as well as mechanical, thermal, and chemical resistances.[5,10,12,13] Dextrans are also widely used for biomedical applications because of their biocompatibility and ease of modification and biodegradation.[14] For the treatment of skin lesions, among various substances, there is a consensus in the use of silver sulfadiazine at 1% (w/w) for the treatment of burns and skin infections. Silver sulfadiazine is effective against a wide microbiota of gram-negative bacteria such as *Escherichia coli*, Enterobacter, Klebsiella, and *Pseudomonas aeruginosa*, including also gram-positive bacteria such as *Staphylococcus aureus* and yeasts such as *Candida albicans*.[15] Silver sulfadiazine is indicated in the treatment and prevention of infections in burns, with classification of anti-infectant agent in the form of cream at 1% (w/w); short-term adjuvant treatment, for infections in both leg ulcers and pressure ulcers; prophylaxis of infection in areas of skin grafts prone to abrasion.[16] Silver sulfadiazine in the form of cream requires multiple daily applications, interfering with the healing process as the wound dressing exposes patients to infectious agents, in addition to the pain and trauma that it causes because the cream is not biodegradable and requires removal before reapplication.[17] Considering the growing importance of the pharmaceutical form hydrogel, the aim of the present research work was to develop and evaluate a hydrogel using PVA and/or dextran as raw materials, containing incorporated silver sulfadiazine, for antimicrobial topical applications. The optimized hydrogel formulation integrating silver sulfadiazine was subsequently fully characterized physicochemically, encompassing determination of pore size and porosity via X-ray tomography, surface morphology via scanning electron microscopy (SEM), thermal analyses via thermogravimetric analysis (TGA), and differential scanning

Correspondence to: Marta M. D. C. Vila (Telephone: +55-15-2101-7181; Fax: +55-15-2101-7000; E-mail: marta.vila@prof.uniso.br)

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calorimetry (DSC), Fourier transform infrared (FTIR) spectrophotometry, and X-ray diffraction (XRD) analyses.

MATERIALS AND METHODS

Materials

Chemicals

All reagents used were of analytical grade or better, and were used without further purification. Tap water was purified in a Milli-Q Elga Purelab system (Molsheim, France) to a final conductivity of approximately 18.2 MΩ cm^{-1}. PVA with a hydrolysis degree of 89% (PVA89), aqueous glutaraldehyde solution (25%, v/v), and PEG 4000 were purchased from Dinâmica Produtos Químicos Ltda (Diadema, São Paulo, Brazil). PVA with a hydrolysis degree of 98.99% (PVA99) was purchased from Sigma–Aldrich (St. Louis, Missouri). Dextran from Amresco Biochemicals Life Research Products was acquired from Sellex (São Paulo, Brazil). Silver sulfadiazine with a purity degree of 100.06%, considering 29.3% silver, was purchased from Valdequímica Produtos Químicos Ltda (São Paulo, Brazil). The ointment with silver sulfadiazine at 1% (w/w), with tradename Dermazine® and manufactured by Silvestre Labs Química Farmacêutica Ltda (Rio de Janeiro, Brazil) was purchased from the local market at the city of Sorocaba (Sorocaba, São Paulo, Brazil).

Biological Materials

The bacterial strain utilized in the antimicrobial assays was *Staphylococcus aureus* CCCD S007, and was acquired from Cefar Diagnóstico Ltda (São Paulo, Brazil). The microbiological growth media utilized were BHI broth (Brain Heart Infusion) from Fluka Analytical (St. Louis, Missouri) and Manitol Salt Agar from Prodimol Biotecnologia S.A. (São Paulo, Brazil).

Analytical Equipment

Fourier transform infrared spectra were gathered in a FTIR Spectrophotometer from Agilent (model Cary 630; Santa Clara, California). X-ray diffractograms were gathered in a X-ray diffractometer from Shimadzu (model XRD7000; Kyoto, Japan). Thermogravimetric characterization of the hydrogels was accomplished via thermogravimetric analysis (TGA), whereas thermal analyses were pursued by DSC. TGA analyses were carried out using a thermogravimeter from TA Instruments (model 2050; New Castle, Delaware), whereas the DSC analyses were carried out using a differential scanning microcalorimeter from TA Instruments (model MDSC 2910). The surface of the hydrogels containing incorporated silver sulfadiazine was duly observed in a scanning electron microscope (model JSM-63660 CV; JEOL, Tokyo, Japan). Hydrogel samples were sputter-coated with colloidal gold under vacuum, and placed in the microscope chamber. Microphotographs were gathered using electron beams with acceleration speeds of 10–20 keV. The samples were randomly scanned and photomicrographed at magnifications of ×50, ×200, and ×1000. A computed tomographer via X-ray transmission from Bruker microCT (model SkyScan 1174; Kontich, Belgium), and an energy-dispersion X-ray fluorescence (EDXRF) spectrometer from Shimadzu (model EDX-700) were utilized in all nondestructive analyses, for gathering tomographic images of the hydrogels. The analysis software utilized for processing the tomographic images were CTVox™ (version 2.6.0 r908–64bit, from Bruker microCT) and CTan™ (version 1.13.5.1–64bit, from Bruker microCT), and CTvol (version 2.2.3.0–64bit, from Bruker microCT).

Experimental Procedures

Preparation of Hydrogels

To produce the hydrogels, aqueous dispersions of PVA99 (10%, w/v), PVA89 (10%, w/v), dextran (15%, w/v), and silver sulfadiazine (10%, w/v) were previously prepared. One-hundred gram PVA (99 or 89) was weighed in an analytical scale, added with 1000 mL ultrapure water and the resulting mixture kept under magnetic stirring (~300 rpm) and heating (~80 ± 5°C) during approximately 2 h, using a magnetic stirring device from Fisaton (model 752 A; São Paulo, Brazil). Following this time period, the mixture was stirred during approximately 6 min at 5200 rpm using an UltraTurrax (model T25D; IKA Werke GmbH and Company KG, Staufen, Germany) homogenizer. Afterwards, the mixture was left under mechanical stirring at 80 ± 5°C for approximately 2 h. The dextran aqueous solution at 15% (w/v) was prepared by adding 150 g dextran with 1000 mL ultrapure water, with mechanical stirring in a magnetic stirring device (~300 rpm) at room temperature (±25°C) during approximately 10 min. The aqueous solution of silver sulfadizine at 10% (w/v) was prepared by dispersing 10 g of silver sulfadiazine in 100 mL of PEG 4000 at room temperature (±25°C). Eleven hydrogel formulations and two cross-linking methodologies were proposed and duly tested, being one a chemical method (using glutaraldehyde as cross-linking agent) and the other a physical one (cryogelification). All hydrogel formulations tested are described in Table 1. PVA dispersions (either PVA99 or PVA89) and dextran were mixed and heated up to 80 ± 5°C under magnetic stirring (~300 rpm) for

Table 1. Formulations Tested for Hydrogels of PVA/Dextran with Silver Sulfadiazine

	Hydrogel Formulation										
Component (%, w/w)	I[a]	II[a]	III[a]	IV	V[a]	VI[a]	VII[a]	VIII[a]	IX	X	XI
PVA99	20	15	10	12.5	15	15	5	–	10	7.5	5
PVA89	–	–	10	–	5	5	15	20	–	–	–
Dextran	–	4.5	–	5.25	–	4.5	4.5	–	6.75	8.25	9.75
PEG 4000	10	10	10	10	10	10	10	10	10	10	10
Silver sulfadiazine	1	1	1	1	1	1	1	1	1	1	1
Ultrapure H_2O, ast	100	100	100	100	100	100	100	100	100	100	100

[a]For the chemical cross-linking process, 5.0 mL of glutaraldehyde aqueous solution at 25% (v/v) were added.
ast, amount sufficient to; PVA99, polyvinyl alcohol with 99% degree of hydrolysis; PVA89, polyvinyl alcohol with 89% degree of hydrolysis.