DENTAL MATERIALS 23 (2007) 977–982

available at www.sciencedirect.com

ScienceDirect

journal homepage: www.intl.elsevierhealth.com/journals/dema

Effects of solubilizing surfactants and loading of antiviral, antimicrobial, and antifungal drugs on their release rates from ethylene vinyl acetate copolymer

Padmavathy Tallury[a], *Marcus K. Randall*[a], *Khin L. Thaw*[a], *John S. Preisser*[b], *Sid Kalachandra*[a,*]

[a] *Center for Oral and Systemic Diseases, Department of Periodontology, School of Dentistry, University of North Carolina, Chapel Hill, NC 27599-7455, USA*

[b] *Department of Biostatistics, School of Public Health, University of North Carolina, Chapel Hill, NC 27599-7455, USA*

ARTICLE INFO

Article history:
Received 24 March 2006
Received in revised form
21 August 2006
Accepted 31 August 2006

Keywords:
Drug delivery
EVA matrix
Nystatin
Surfactants
Chlorhexidine diacetate
Acyclovir
Drug loading

ABSTRACT

Objectives. This study investigates the effects of surfactants and drug loading on the drug release rate from ethylene vinyl acetate (EVA) copolymer. The release rate of nystatin from EVA was studied with addition of non-ionic surfactants Tween 60 and Cremophor RH 40. In addition, the effect of increasing drug load on the release rates of nystatin, chlorhexidine diacetate and acyclovir is also presented.

Method. Polymer casting solutions were prepared by stirring EVA copolymer and nystatin (2.5 wt.%) in dichloromethane. Nystatin and surfactants were added in ratios of (1:1), (1:2) and (1:3). Drug loading was studied with 2.5, 5.0, 7.5, and 10.0 wt.% proportions of nystatin, chlorhexidine diacetate and acyclovir incorporated into a separate polymer. Three drug loaded polymer square films (3 cm × 3 cm × 0.08 cm) were cut from dry films to follow the kinetics of drug release at 37 °C. Ten milliliters of either distilled water or PBS was used as the extracting medium that was replaced daily. PBS was used for nystatin release with addition of surfactants and water was used for the study on drug loading and surfactant release. The rate of drug release was measured by UV-spectrophotometer. The amount of surfactant released was determined by HPLC.

Results. The release of nystatin was low in PBS and its release rate increased with the addition of surfactants. Also, increasing surfactant concentrations resulted in increased drug release rates. The release rates of chlorhexidine diacetate ($p < 0.0001$), acyclovir ($p < 0.0003$) and nystatin ($p < 0.0017$) linearly increased with increasing drug loads. The amount of surfactants released was above the CMC.

Significance. This study demonstrates that the three therapeutic agents show a sustained rate of drug release from EVA copolymer over extended periods of time. Nystatin release in PBS is low owing to its poor solubility. Its release rate is enhanced by addition of surfactants and increasing the drug load as well.

* *Corresponding author.* Tel.: +1 919 966 4545; fax: +1 919 966 7537.
E-mail address: sid_kalachandra@dentistry.unc.edu (S. Kalachandra).

0109-5641/$ – see front matter
doi:10.1016/j.dental.2006.08.005

1. Introduction

Nystatin is an antifungal drug that is widely used for treating oral infections. It has low solubility in water and saliva [1]. It is known that the solubility of sparingly water-soluble drugs can be increased through the addition of surfactants [2]. Solubilization of water insoluble drugs by micelles has long been investigated as a means of improving solubility for drug delivery [3] and the incorporation of a wide variety of drugs into micelles formed from a large variety of surfactants in particular non-ionic surfactants have been studied [4,5]. Nystatin release from a chewing gum formulation as drug delivery device with addition of non-ionic surfactants Tween 60 (polyoxyethylene sorbitan mono stearate), Cremophor RH 40 (polyoxyl 40 hydrogenated castor oil) and Panodan AB 90 (diacetyl tartaric acid esters of mono and diglycerides of vegetable fats) was studied by Andersen et al. [6]. The addition of surfactants promoted a far higher release of nystatin. There are other reports that have demonstrated an increase in drug release due to the addition of surfactants [7–10].

The main objective of this study is to investigate the effect of surfactants Tween 60 (Tween) and Cremophor RH 40 (Cremophor) on the release rate of nystatin from ethylene vinyl acetate (EVA) copolymer. Our second objective is to study the influence of drug loading on the rate of release of nystatin, chlorhexidine diacetate and acyclovir from EVA. Our previous studies have shown the ability of EVA, a biocompatible copolymer to deliver drugs at constant concentrations for an extended period of time [11–15]. This copolymer system is able to release the drug over several weeks and therefore may be useful as a drug carrier in the treatment of oral infections.

2. Materials and methods

The materials used in this study are detailed in Table 1.

2.1. Preparation of thin polymer films

Drug loaded EVA polymer films were prepared by solvent evaporation technique according to our earlier studies [11–15]. Nystatin loaded samples with surfactants Tween and Cremophor were prepared similarly in drug to surfactant ratios of (1:1), (1:2) and (1:3). For the experiment on the effect of surfactants on nystatin release, nystatin incorporated was 2.5 wt.% of EVA. The release rates from these samples were compared to those of nystatin alone in order to study the effect of surfactants. In addition, drug loading was studied with 2.5, 5.0, 7.5, and 10.0 wt.% proportions of nystatin, chlorhexidine diacetate, and acyclovir incorporated into a separate polymer.

Table 1 – Materials and suppliers

Material	Suppliers
Chlorhexidine diacetate	Sigma–Aldrich
Nystatin	Sigma–Aldrich
Acyclovir	Sigma–Aldrich
Ethylene vinyl acetate ((Elvax 40 Wg) (EVA))	DuPont
Dichoromethane	Mallinckrodt Baker Inc.
Phosphate buffer saline (PBS)	Sigma–Aldrich
Tween 60 (Tween)	Sigma–Aldrich
Cremophor RH 40 (Cremophor)	Bayer

2.2. Determination of release rate

Three drug loaded polymer square films (3 cm × 3 cm × 0.08 cm) were cut from dry films to follow the kinetics of drug release at 37 °C. Ten milliliters of either distilled water or PBS was used as the extracting medium. The release of nystatin with surfactants was studied in PBS while water was used for the release study of increasing loads of chlorhexidine diacetate, acyclovir and nystatin. Fresh 10 ml samples of the media were used daily for 12–14 days and the extracts were analyzed for a decrease in concentration by measuring the optical density (OD) spectrophotometrically (Hitachi U-2810 Spectrophotometer) at wavelengths (λ_{max}) where the maximum absorption occurred. The λ_{max} values were 306, 257.5 and 253 nm for nystatin, chlorhexidine diacetate and acyclovir, respectively. Using standard plots of OD versus concentration, the drug concentration was determined each day.

UV spectral measurements were also made for the two surfactants Tween and Cremophor. The surfactants did not exhibit any absorbance in the region 200–400 nm and did not interfere with the determination of absorbance values for nystatin. Also, the standard plots of nystatin were similar with and without the addition of surfactants.

The amount of Tween and Cremophor released from the nystatin loaded EVA films in water at 37 °C was determined by high performance liquid chromatography (HPLC). The used HPLC set up was equipped with a Waters Atlantis dC18 3.9 mm × 150 mm, a Waters 2695 separation module, a Waters 2420 evaporative light scattering detector with settings 75% nebulizer heater level; 80 °C drift tube temperature; 20 gain; 30 psi nitrogen gas. The mobile phase was a mixture of A (85% Fisher HPLC Grade Water) and B (15% Fisher HPLC Grade acetonitrile) with a gradient elution of 70% A and 30%B for the first 5 min and 85% A and 15% B for the next 11 min. The flow rate was 1.00 ml/min and sample injection was 20 μl. Using standard surfactant plots, the concentration of the surfactant released from the EVA films was measured.

2.3. Determination of solubility of drugs

Solubility of the three drugs chlorhexidine diacetate, acyclovir and nystatin was determined individually in 5 ml of water by stirring a saturated solution of the drug at 37 °C for 24 h. The excess drug was filtered and the solution was analyzed by UV spectrophotomer.

2.4. Statistical analysis

For each study, one-way analysis of variance was applied to the drug release rates transformed to the log scale to achieve approximate normality and variance homogeneity. If the overall *F*-test for drug with/without surfactant was statistically significant at the 0.05 level, nine post hoc contrasts were tested. Six of these comparisons involved the assessment of