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Composite hydrogels as a vehicle for releasing drugs with a wide range of hydrophobicities

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ABSTRACT

Many vitamins, bioactive lipids and over 40% of newly developed drugs are hydrophobic, and their poor water solubility limits their delivery using conventional formulations. In this work we investigated a composite gel system formulated from microemulsions embedded in alginate hydrogels, and showed that it is capable of loading several hydrophobic compounds with a wide range of aqueous solubility. All gels were clear, with no precipitations, indicating the solubility of the drugs in the gels. The release behavior was similar for different microemulsion formulations, various drugs and increasing concentrations of a drug. These findings indicate that our system could potentially act as a generic system, where the properties of the release do not depend on the drug but rather on the attributes of the gel. The structure of composite gels was investigated using small-angle scattering of X-rays and neutrons (SAXS and SANS, respectively). SANS showed more sensitivity to the structure of the microemulsion in the composite gel than SAXS did. SAXS and SANS plots of the composite gels show that both the droplets and the gel network preserve their structure when mixed together.

1. Introduction

Hydrogels, hydrophilic polymer networks capable of absorbing up to thousands of times their dry weight in water, have numerous uses in pharmaceutical and medical fields. Applications span from tissue engineering [1–4], wound dressings [5] and tissue adhesives [6] to drug delivery [7,8] and more. In drug delivery, hydrogels are classically employed to deliver small hydrophilic drugs [9]. To date, hydrophobic drugs are inefficiently loaded and released from hydrogels, due to the high water content and the hydrophilic nature of the polymer.

Poorly water-soluble compounds that promote health are very common. These include vitamins, (such as vitamins A and D), antioxidants (curcumin, carotenes), bioactive lipids (omega-3, omega-6), peptides and drugs. It is estimated that more than 40% of newly developed drugs are extremely hydrophobic, and this number is steadily increasing [10,11]. Different strategies are being developed in order to exploit the advantages of hydrogels and implement them for use with lipophilic compounds. One strategy is to incorporate hydrophobic domains into the polymer [12]. Another is to introduce nanoparticles that encapsulate hydrophobic compounds. Cyclodextrins can be used to stabilize hydrophobic compounds, for example in alginate foams [13] and dextran-based hydrogels [14]. In one study, cationic nanoparticles were combined with Pluronic® F127 hydrogel and their delivery of hydrophobic anticancer drugs to the bladder was examined [15]. In another study, the hydrophobic agent fluorescein was released from polymethyl methacrylate nanoparticles dispersed in pH-responsive hydrogels [16]. Jagadeesan et al. [17] studied the delivery of polar and non-polar molecules by microgels produced by a microfluidics method. We recently designed a composite hydrogel, where a microemulsion is dispersed in a crosslinked polysaccharide [18]. An oil-in-water microemulsion is a dispersion of oil nanodroplets in water stabilized by an interfacial film of amphiphilic molecules. Lipophilic drugs can be incorporated in the core of the droplets.

Many studies focus on the release of one hydrophobic drug, while only a few compare the release patterns of different hydrophobic drugs from the same hydrogel system. Most of the latter studies found a relation between the chemical attributes of the drug and the release rate. Two hydrophobic drugs, benzophenone and tamoxifen, exhibited different release patterns from self-assembling gels containing cyclodextrins (CD) [19]. Another study on hydrogels having β-CD functionality showed that the release rate depends on the inclusion complex formation capability of β-CD moieties and hence varies from drug to drug [20]. Jagadeesan

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et al. [17] reported on microgels incorporating drugs with a log*P* range between 1.3 and 2.8. Each drug displayed a different flux, which seemed to be linked to the hydrophobicity of the compounds and hence to their affinity to the interior of the microgel particles. Inoue et al. [21] showed that the release rate from a hydrophobically modified polyelectrolyte depends on the hydrophobicity of the drug. Different release rates were also observed in a hydrogel based on nanoparticles [22], and niosomes embedded in locust bean gum and xanthan hydrogel [23]. In contrast, similar diffusion coefficients were observed for several hydrophilic and hydrophobic drugs released from bovine serum albumin (BSA) and polyethylene glycol (PEG) hydrogels [24]. An explanation as to why diffusion coefficients were independent of the water solubility was not provided.

Designing a hydrogel delivery system in which the release rate is less sensitive to the chemical nature of the drug may be beneficial in some cases. The requirement for a specific drug is determined by the clinical situation, and therefore properties such as hydrophobicity cannot be easily altered. On the other hand, the hydrogel's properties such as mesh size can be manipulated to provide an easy means to control the release pattern. We suggest that a system composed of microemulsions embedded in a hydrogel could meet the above requirements. Microemulsions are dynamic in nature; the time between interdroplet exchanges is in the range of microseconds to milliseconds [25]. In addition, their size is comparable to or smaller than the mesh size of the gel [26]. Thus, we hypothesize that the properties of the gel are more significant during the release process than those of the droplets.

In this work we fabricated composite gels incorporating different microemulsion formulations, and examined the ability of these gels to solubilize several hydrophobic entities. The term composite hydrogels can refer to various types of hydrogels, such as "plum pudding" hydrogel networks which contain particulate systems [9] or a combination of polymers [27]. Our hydrogel is composed of alginate, a biocompatible water-soluble polysaccharide, crosslinked with calcium cations. We explored the mechanism of release of these compounds from different composite gels, and studied which components of the system contribute to the release patterns. The structure of the microemulsion and the composite gel were studied using small-angle scattering, with X-ray (SAXS) and neutron (SANS) radiation. Understanding the system's structure and its release behavior is a step towards tailoring similar composite gels for many more hydrophobic compounds.

2. Materials and methods

2.1. Materials

D(+)-Gluconic acid δ-lactone (GDL), ethylene glycol-bis(2-aminoethylether)-N,N,N′,N′-tetraacetic acid (EGTA), sorbitan laurate (Span 20) and isopropyl myristate (IPM) were purchased from Fluka. $CaCl_2$ was purchased from J.T. Baker, and Polysorbate 80 (Tween 80) from Merck. Isopropyl palmitate (IPP), cholecalciferol (vitamin D_3), progesterone, ketoprofen and Phenoxazon-9 (Nile red) were obtained from Sigma. Alginate (LF 200 FTS) was supplied by FMC Biopolymers (Drammen, Norway). All materials were used as received.

2.2. Microemulsion preparation

The microemulsion was prepared by mixing the surfactants Tween-80 and Span-20 or Span-80 with oil (IPM or IPP), followed by dropwise addition of double-distilled water. In order to obtain a clear microemulsion, the solution was allowed to equilibrate for 24 h. Drug-containing microemulsions were prepared by adding the appropriate amount of drug to the final microemulsion. Concentrations of drug in the microemulsion were 10 mg ml^{-1} for ketoprofen, 2 mg ml^{-1} for vitamin D_3 and progesterone, and 0.5 or 1 mg ml^{-1} for Nile red. These concentrations were the highest concentration of each drug where no precipitations were observed visually.

2.3. Size of droplets

The average particle size (volume average size) was measured by dynamic light scattering (Nano ZS zetasizer, Malvern Instruments Ltd, UK) at 25 °C with an He–Ne laser (633 nm) at an angle of 173°.

2.4. Gel preparation

Alginate was dissolved in double-distilled water; thereafter, the microemulsion was added and stirred with a magnetic stirrer. The microemulsion concentration in the gel was 10% wt./vol. A calcium source in the form of a pre-prepared Ca–EGTA solution was introduced next, followed by a fresh GDL solution. GDL induces slow release of calcium ions from the Ca–EGTA complex, allowing gelling of the alginate solution [28]. The Ca^{2+}:GDL molar ratio was 1:2. For the preparation of the Ca–EGTA solution, equal molar amounts of $CaCl_2$ and EGTA were dissolved in water and the pH adjusted to 7 by adding 1 M NaOH. Drug release measurements were done at least 24 h after GDL addition, to allow for the alginate solution to gel completely [28]. For $CaCO_3$, a molar ratio of 1:2 guarantees only slightly acidic acid in the final hydrogel, thus the presence of acidic gel is negligible [29]. We chose this ratio arbitrarily in our experiment for Ca–EGTA, and this parameter was constant across all of our experiments. The molar ratio could have an effect on the final structure of the gel. Unless stated otherwise, final compositions were 10 mg ml^{-1} alginate and 20 mM calcium. Gels were casted by pouring 600 μl of the solution into a ring mold with a diameter of 1.4 cm.

2.5. Drug release

A 6 ml volume of double-distilled water was added to 0.6 ml of gel. The samples were placed in a 37 °C bath and shaken at a rate of 100 rpm. At each time interval, 0.2 ml of the surrounding medium was sampled and replaced with 0.2 ml of fresh water. The sample was measured in a spectrophotometer at a wavelength of 233 nm for the microemulsion and 535 nm for Nile red. Three types of crosslinked alginate gels were prepared: (i) without drug or microemulsion (control), (ii) with drug-free microemulsion and (iii) with microemulsion containing a drug. Four samples from each type of gel were prepared. The control gel was used as blank, and drug or microemulsion concentrations were calculated from calibration curves. UV spectroscopy was carried out on a 96-well plate with a Synergy HT microplate reader (Bio-Tek Instruments, Winooski, VT, USA). Results present an average of four gels.

2.6. Swelling experiments

Double-distilled water was added to 0.6 ml of gel. The samples were placed in a 37 °C bath and shaken at a rate of 100 rpm. The percentage of swelling was calculated by the weight of the gel at each time point normalized by its initial weight. Five gels were averaged to give the final percentage of swelling.

2.7. Small-angle X-ray scattering

X-ray scattering was performed using a small-angle diffractometer (a Molecular Metrology SAXS system with Cu K_α radiation