Controlling surface porosity and release from hydrogels using a colloidal particle coating

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Abstract

Recent studies suggest that coating microcapsules by a shell composed of impenetrable colloidal particles (thereby forming ‘colloidosomes’) can be used to control surface porosity, and therefore, permeability. The voids between the particles in the coating define the size of the surface pores available for transport. However, to date, data demonstrating this selectivity has been largely qualitative. In this paper we examine, quantitatively, the effect of a surface coating (shell), composed of colloidal particles, on release from hydrogels. We find that the presence of a colloidal shell does indeed reduce the rate of transport of three model molecules: Aspirin, caffeine, and FITC-dextran with MW of ~3000–5000. Contrary to expectation, however, we find that for all three molecules the reduction in transport rate is largely independent of the dimensions of the particles composing the shell, despite differences that range over three orders of magnitude. In the case of the small molecules, caffeine and aspirin, the colloidal shell reduces the effective diffusion coefficient by a factor of 3. In the case of dextran, the suppression in the release rate due to the colloidal shell was much larger. These results are explained using a simple diffusion model that accounts for the volume fraction and diameter of the colloidal particles in the shell, and the size of the diffusing molecules.

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1. Introduction

The ability to control the rate of transport out of and into polymer films or particles is required in various applications [1–3]. Industries such as pharmaceuticals, cosmetics, food and household cleaners need the ability to design the release profile of an entrapped material such as a drug [2,4–6] or a fragrance [7,8]. Other utilizations require inhibition of transport, such as water loss and gas permeation to food products [9] or immuno-agents into biomedical implants [10–13].

Various methodologies have been developed to control the transport into and out of polymeric particles. Many rely on the polymer properties (e.g. degradable, glassy or crystalline) to control transport [1,14,15]. Others use a surface film to control properties such as chemistry, charge or roughness, thereby affecting transport [9,16,17].

An acute challenge that has not been successfully addressed by current coatings is the ability to selectively control the transport of different components. For example, an ideal coating for fresh fruit would inhibit water loss, while allowing selective transport of gases to slow the aging process [9]. In encapsulated cells that are implanted as a therapy for diseases such as Type I Diabetes, transport of nutrients and active components (e.g. insulin) must not be hindered, while that of immuno-system agents must be suppressed [11,12,18–22]. One way of obtaining such selectivity is by controlling surface porosity, namely, the dimensions of the pores through which transport of the diffusants occurs. Components that are smaller than the pores can cross the surface freely, while that of larger molecules is suppressed.

Colloidosomes (a term coined by Dinsmore et al. [23]) are microcapsules characterized by a coating, or shell composed of self-assembled colloidal particles (see Fig. 1). Like their lipid-based and polymer-based counterparts, colloidosomes can encapsulate aqueous volumes and have a wide range of applications in bio-related technologies as well as other industries including food and cosmetics [2,6,22–25]. The colloidal shell consists of uniform particles that can range in size from nanometers to microns. As a result, the surface porosity, namely, the size of the ‘voids’ or surface pores can range over similar orders of magnitude.

In highly packed shells, the porosity of colloidosome shells has been reported to vary as a function of the size of the colloidal particles. Lee and Weitz [26] find, qualitatively, that a small molecule can diffuse through a colloidosome shell composed of particles on the order of 10–20 nm, but a high MW polymer with a radius of gyration on the order of 40 nm cannot. These observations are consistent with the fact that the size of the pores between the closely packed particles in the colloidosome shell is expected to be of...
order 10% of the particle radius [27], allowing the small molecule (with a radius of ~1 nm) to diffuse, but obstructing the transport of the large polymer. In a more detailed study, Kim et al. [25] found that the permeability of fluorescein through a colloidal shell composed of 1 μm particles coating a microgel drop was only slightly reduced when compared to the uncoated microgel. However, a shell composed of 80 nm colloidal particles was found to suppress the rate of permeation by an order of magnitude [25].

The Lee and Weitz [26] and Kim et al. [25] papers suggest that densely packed shells composed of colloidal particles may be used to suppress transport of materials through the shell. However, the correlation between the magnitude of the transport suppression (complete inhibition versus a slowing down of the diffusion rate), the size of the colloidal particles composing the shell and their packing density, and the dimension (or MW) of the diffusing molecules has not been established.

In this paper we examine, systematically, the transport of model molecules through a self-assembled colloidal particle shell. Due to the self-assembly synthesis methodology, the packing density of the particles is moderate, when compared to the closely-packed systems studied by Lee and Weitz [26] and Kim et al. [25]. We use alginate gels as the scaffold for the colloidal shell, since this biocompatible polymer is commonly used in relevant biomedical and food applications [28–30]. Also, to enable systematic measurements of the flux through the shell, we measure the release of molecules from a flat film, rather than colloidosome microgels. This enables circumvention of issues that may arise in the microgel system, such as encapsulation inhomogeneity or gel polydispersity.

We compare shells composed of different particle sizes ranging from 20 nm to 3.3 μm. Caffeine and acetylsalicylic acid (aspirin) were used as models for relatively small (MW ~200 g/mol) hydrophilic molecules, and dextran (MW 3000–5000) as a model for larger molecules.

It might have been expected that, due to the moderate packing density in our shells (and the corresponding larger size of the voids, or pores, in the shell) that the colloidal coating would not significantly affect transport. However, our results show that the presence of the loosely-packed colloidal shell does indeed reduce the rate of transport for both types of molecules. Contrary to previous studies, however, we find that for the three molecules, the reduction in transport rate is largely independent of the dimensions of the particles composing the shell, despite differences that range over three orders of magnitude in the colloidal particle diameter. In the case of the small molecules, caffeine and aspirin, the initial release rate from a coated film is approximately one-third that of the release from the uncoated films, and 100% release required twice the amount of time. This observation is in excellent agreement with the predictions of a theoretical model for transport through composite media.

In the case of dextran, the suppression in the release rate due to the colloidal shell was much more dramatic: The initial release rate from the coated films was an order of magnitude lower than from the uncoated ones, and only approximately 10% of the molecules was released from the coated films over the time needed for 100% release from uncoated ones. This enhanced suppression is attributed to a specific chemical interaction between dextran and the polystyrene particles, leading to dextran adsorption onto the particles in the colloidal shell. Therefore, this result cannot be readily generalized to other large molecules.

Our results enable evaluation of the effect of a colloidal shell on the transport of a given molecule as a function of the colloidal particle size and the characteristics of the molecules of interest.

2. Materials and methods

2.1. Materials

Sodium alginate, calcium chloride, FITC-Dextran (average MW 3000–5000), aspirin and caffeine were purchased from Sigma–Aldrich (St. Louis, MO and Milwaukee, WI, USA). Amidine (C(NH2)=NH+) functionalized polystyrene (PS) particles (sizes 20 nm–3.3 μm) were purchased from Invitrogen Molecular Probes. All reagents were used as purchased with no further purification.

2.2. Synthesis of alginate films

2.2.1. Uncoated films

A 2% w/v sodium alginate in water solution was shaken and sonicated to remove any excess air. One milligram of a model molecule was dissolved in 400 μL of the sodium alginate solution and placed in a 30 mL (25 mm × 95 mm) glass vial. The vials were placed in a convection oven at 40 °C overnight to evaporate the excess solvent. Five-hundred microlitres of a 10 wt.% solution of calcium chloride in water was added to the alginate and allowed to crosslink for 5 min. Excess calcium chloride was removed by washing with deionized water.

2.2.2. Coated films

A 4% w/v PS particle solution (0.5 mL) was added to the top of the uncoated hydrogel films and incubated overnight. The films were subsequently washed with deionized water to remove excess, unadsorbed, particles. The resulting colloidal coating was found to be loosely packed. Using micrographs (not shown), the average particle packing density, defined as the area occupied by particles over the entire surface area, was found to be of order 30%, independent of particle size (note that close-packing is of order 60–75%).

2.3. In vitro assay

For each study, five samples of the same type were used for reproducibility purposes. Each vial contained 20 mL of deionized water. All vials and samples were kept in a dark environment as fluorescently labeled dextran is light-sensitive. Aliquots of 1 mL were taken at structured time periods from each vial to measure ultraviolet absorbance for caffeine and fluorescence emission.
spectra for FITC-dextran using spectrometers. One millilitre of pure deionized water was replaced each time a sample was taken and the amount of molecule removed in each sample was accounted for.

2.4. Measurement techniques

2.4.1. Ultraviolet spectroscopy

Concentrations of molecules released were calculated using a linear calibration curve. Four milligrams of caffeine were dissolved in 10 mL of deionized water. The solution was shaken for 2 min and diluted by half. The dilution was repeated until eight samples were made with pure water as the lowest concentration. The absorption spectra showed peaks at 273 nm, where all further measurements were taken. Concentrations were in the linear regime to minimize error.

2.4.2. Fluorescence spectroscopy

At low concentrations of FITC-dextran, there is a linear relationship between fluorescence intensity and concentration. All samples were kept in this linear regime. Calibration samples were made with the same procedure as caffeine. The excitation wavelength was kept at 493 nm and emission spectra were measured three times for each sample in the range of 505–525 nm and an integration time of 0.25 s. Emission peaks were typically measured at 513 nm.

3. Results and discussion

3.1. Release studies

3.1.1. Uncoated films

We first examine the release of the three model molecules (caffeine, aspirin and FITC-dextran) from the uncoated films. As shown in Fig. 2, all three molecules seem to follow a classic release profile composed of a relatively linear initial stage, followed by a reduced rate until 100% of the molecule is released [31]. We see that 100% release of aspirin occurs much faster than caffeine, and dextran takes the longest time. We expect that the rate of release would depend on the diffusion coefficient of the model molecules in the alginate film. In the case of the smaller molecules, this should largely correspond to their diffusion coefficient in water, since our films are highly swollen. Unfortunately, we are not aware of a reported value for the diffusion coefficient for aspirin in water; however, a previous study utilized an approximation the diffusion coefficient of o-aminobenzoic acid in water, which is $8.6 \times 10^{-6} \text{cm}^2/\text{s}$ [32]. This is slightly faster than the diffusion coefficient of caffeine in water, $6.3 \times 10^{-6} \text{cm}^2/\text{s}$ [33], and is in agreement with the fact that aspirin achieves 100% release somewhat faster than caffeine, but within the same order of magnitude period.

In the case of dextran, the diffusion coefficient in water is sensitive to its MW and its functionalization. Literature values suggest that FITC-dextran with MW similar to ours, in water, should have a diffusion coefficient of order $10^{-7} \text{cm}^2/\text{s}$ [34–37]. However, this value would be reduced when the media contains polymer chains [35]. Thus, our finding that dextran requires a period of time for 100% release that is more than an order of magnitude slower than that of the small molecules (~144 h, or ~6 days), is reasonable.

The rate of release from a flat film was derived by Crank [31] to be

$$f = 1 - \frac{M(t)}{M_0} = \sum_{n=0}^{\infty} \frac{8}{\pi^2(2n+1)^2} \exp \left[ -\pi^2 \left( n + \frac{1}{2} \right)^2 \frac{D}{h^2} t \right]$$

(1.a)

which may be approximated by

$$f \approx 1 - \exp \left[ -\frac{D}{h^2} t \right]$$

(1.b)

where $f$ is the fraction of molecules released at time $t$, $M$ is the mass of the molecule in the film ($M_0$ is the initial amount), $h$ is the thickness of the film and $D$ is the diffusion coefficient in the film.

Using Eq. (1.b) to fit the aspirin, caffeine and dextran release profiles (Fig. 3), we see that the model fits aspirin and dextran very well, but in the case of caffeine, the fit is less accurate. The values of the fit coefficient, $D/h^2$, used for the release from the uncoated films in Fig. 3 are $\sim1 \times 10^{-3} \text{cm}^2/\text{s}$ for aspirin, $3 \times 10^{-4} \text{cm}^2/\text{s}$ for caffeine, and $5 \times 10^{-6} \text{cm}^2/\text{s}$ for dextran. Taking our film thickness to be on the order of 0.1 cm, the model fit yields values for the diffusion coefficients of order $10^{-5} \text{cm}^2/\text{s}$ for aspirin, $10^{-6} \text{cm}^2/\text{s}$ for caffeine and $10^{-8} \text{cm}^2/\text{s}$ for dextran. Considering the significant approximations involved and the sensitivity to the (unmeasured) value of the film thickness due to the scaling of the fit coefficient as $h^2$, these values are in relatively good agreement with the published diffusion coefficient values for all three molecules [32–36].

The results presented in Fig. 3 demonstrate that the release of the model molecules from alginate films generally follow expectations from classic diffusion models [31]. Therefore, we can now examine the effect of the colloidal shell, or coating, on the release.

In Fig. 4, we compare the release of each of the model molecules from uncoated alginate films to that from films coated by colloidal particles. We see that, in all three systems, the release from the uncoated films is much more rapid than the release from films coated by the colloidal shells. Moreover, contrary to the findings of Kim et al. [25], the size of the colloidal particles composing the shell does not seem to affect the rate of release (except for dextran transport through the 20 nm particle shell, which is discussed in detail below).

![Fig. 2. Release profiles of aspirin, caffeine, and dextran from uncoated films.](image-url)
In Table 1 we compare features of the release profile for the coated and uncoated films, for the three molecules. The results presented in Fig. 4 and Table 1 are surprising, since it may be expected that the reduction in transport rate would scale

### Table 1

<table>
<thead>
<tr>
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<th>Aspirin</th>
<th>Caffeine</th>
<th>Dextran</th>
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<tbody>
<tr>
<td></td>
<td>Uncoated</td>
<td>Coated(^a)</td>
<td>Uncoated</td>
</tr>
<tr>
<td>Initial release rate (1/h)(^b)</td>
<td>1.64</td>
<td>0.81</td>
<td>1.06</td>
</tr>
<tr>
<td>Approx. time for 100% release (h)</td>
<td>2</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>(D/h^2) (1/s)</td>
<td>(1 \times 10^{-3})</td>
<td>(3 \times 10^{-4})</td>
<td>(3 \times 10^{-4})</td>
</tr>
<tr>
<td>(D^*)</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
</tr>
</tbody>
</table>

\(^a\) Since the release rate is identical for all particle sizes (see Fig. 5), this applies to all coated systems.

\(^b\) The release rate is defined as the fraction of molecules released divided by the time period over which that release occurred (namely, \(\Delta f/\Delta t\)). The initial release rate was evaluated for the period from \(t = 0\) to 30 min for caffeine and aspirin, and \(t = 8\) h for dextran.

\(^c\) Calculated by fitting the diffusion data to Eq. (1).

\(^d\) The ratio between the diffusion coefficient (calculated from Eq. (1)) in the coated system and the uncoated system.

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Fig. 3. Fitting the release of aspirin, caffeine and dextran from uncoated films to Eq. (1,b). The optimal values of the fit parameter \(D/h^2\) are approximately \(1 \times 10^{-3}\) 1/s for aspirin, \(3 \times 10^{-4}\) 1/s for caffeine, and \(5 \times 10^{-6}\) for dextran.

Fig. 4. Release of caffeine (a), aspirin (b) and dextran (c) from uncoated and coated alginate films (d) release of dextran from coated alginate films.
with the size of the colloidal particles composing the shell. However, to understand this behavior it is necessary to consider the mechanism leading to the reduction in transport. The dimensions of the colloidal particles composing the shell are, in all cases, much larger than that of the molecules that diffuse out of the alginate gel (except for dextran and the 20 nm particles). Thus, the suppression must be due to the reduction in the surface area available for transport, since caffeine, aspirin and dextran cannot penetrate through the glassy PS particles used for the shell. However, the close-packed density of spheres (whether in two or three dimensions) is independent of the particle size, so that the area available for transport should be independent of the particle size as observed (see Fig. 4).

This qualitative explanation is supported by theoretical analysis. Several approaches have been derived to examine molecular diffusion through composite media [31,38]. Here we apply the method developed by Bell and Crank [31,39] for transport through porous media, where we approximate the colloidal shell as an ordered array of regions that are not available for transport (the particles) and those that are (the spaces between particles). The effective diffusion coefficient for the composite, $D^*$, may be written as [31,39]

$$\frac{D'}{D} = \frac{1}{1 + \frac{\phi}{a}}$$

(2)

where $D$ is the diffusion coefficient of the molecule in the ‘open’ penetrable regions (in our case, the gel/water areas), $\phi$ is the volume fraction of the impenetrable phase in the coating (namely, the colloidal particles), and $a$ is a geometric parameter that is equal to, in our system, the diameter of the colloidal particle divided by the center-to-center distance between neighboring particles. Thus, for a closely packed system, $a = 1$, while in a system of spherical particles that are not closely packed, $a \approx (6\phi/\pi)^{1/3}$.

As noted earlier, in closely-packed systems, $\phi$ and $a$ are independent of particle size (see, for example, [27]). Eq. (2) suggests that for this case where $a = 1$ and $\phi$ is of order 0.6, the ratio between the diffusion coefficient in the coated and uncoated films should be of order 0.36–0.4. In systems like ours, where the packing density is lower, $D'/D$ is somewhat higher (approximately 0.6).

In Fig. 5, we fit Eq. (1.b) to the release data of caffeine and aspirin from coated films, to obtain values for the effective diffusion coefficient $D^*$ in these systems. We find that the colloidal layer reduces the value of the diffusion coefficient for aspirin and caffeine by a factor of approximately 3 when compared to the uncoated values (Table 1), so that $D'/D = 0.33$ when compared to the value of 0.6 predicted by Eq. (2). However, considering the various approximations (e.g., taking the coated gel to be one volume with a diffusion coefficient $D_c$, rather than accounting for the gel and the shell separately) this factor 2 difference is reasonable. A more detailed model, which is outside the scope of this paper, provides excellent agreement with our data [40].

The effect of the colloidal shell on the diffusion of dextran, however, is quite different when compared to the small molecules. As observed for aspirin and caffeine, the release of dextran is similar for films coated by 1 µm and 3.3 µm. However, the suppression of the release rate is much higher than observed for the small molecules or predicted by Eq. (2). The reduction in both the initial rate and the effective diffusion coefficient, $D'/D$, is by an order of magnitude, rather than a factor of 3. Moreover, transport is completely suppressed when the particles composing the shell are 20 nm. These differences suggest that, in the case of dextran, there is an additional mechanism that reduces or inhibits diffusion through the colloidal shell. The most likely possibility is that, due to its

![Fig. 5](image-url)

Fig. 5. Fitting the release of caffeine (a) aspirin (b) and dextran (c) from coated films to Eq. (1.b). The optimal values of the fit parameter $D'/D^2$ are approximately $3 \times 10^{-4}$ for aspirin, $9 \times 10^{-5}$ for caffeine, and $9 \times 10^{-4}$ for dextran.
higher MW (and consequently larger molecular dimensions), dextran is excluded from the pores between adsorbed particles. Indeed, Kim et al. [25] and Lee and Weitz [26] suggest that size exclusion is the mechanism by which colloidosome shells can be used to selectively inhibit transport of components.

Beck and Shultz [41] found an empirical correlation for the effect of pore size on diffusive transport, given by

$$\frac{D_p}{D} \approx \left[ 1 - \frac{R_m}{R_p} \right]^{-4}$$

(3)

where $D_p$ is the diffusion coefficient of the molecule through the pore, $D$ is the diffusion coefficient of the molecule in the liquid media, $R_m$ is the molecular radius, and $R_p$ is the pore radius. Combining this with the suppression due to the shell (Eq. (2)) leads to the following relationship

$$\frac{D^*}{D} = \left( 1 + \frac{R_m}{R_p} \right)^{-4}$$

(4)

Note that, as long as $R_m/R_p \ll 1$, the effective diffusion coefficient is practically independent of the pore size, and thus of particle dimension, in agreement with the Bell and Crank model presented in Eq. (2) [31]. However, in systems where the pore size is similar to that of the molecular dimensions, additional suppression of the transport would occur.

To utilize Eq. (4), we need to know the characteristic pore size and the dimensions of the diffusing molecule. The molecular dimension of our FITC-dextran ($R_m = 2$ nm) [37,42], is nearly an order of magnitude larger than the dimensions of caffeine (0.376 nm) [43] and aspirin (0.41 nm) [32]. In highly packed colloidal systems, the simulations of Rintoul and Torquato [27] show that the characteristic pore size may be much smaller, 5–10% of the particle radius. However, in systems such as ours where the packing is not necessarily very dense and the pore size is similar to the particle dimensions, the pore size even in the shells composed of the smallest, 20 nm, is larger than the dextran molecular dimensions. Thus, we would not expect molecular size-based suppression in dextran transport, when compared to caffeine and aspirin.

Another potential explanation for the reduction in dextran transport may be the alginate gel. However, since we compare $D^*$ to $D$ in the uncoated gel (rather than to the diffusion coefficient of dextran in water), the suppression must be attributed to the presence of the colloidal shell.

The only potential explanation for the significant suppression in dextran transport through the colloidal shell must therefore be attributed to some specific chemical interaction between the dextran and the particles, which inhibits transport. Indeed, Osterberg et al. [44] found that polysaccharides, including dextran, adsorb readily on PS surfaces. To examine the applicability of this scenario we incubated a mixture of 3.3 µm PS particles and FITC-dextran in water for several hours, followed by repeated centrifugation and washing to remove any unadsorbed dextran. Fluorescence images of the particles (see Fig. 6) show that indeed there is dextran adsorbed to the surface of the particles. Thus, the reduction in dextran transport through the colloidal shell is not due to a mechanical inhibition (namely, the presence of pores defined by the voids between the shell particles) but to a chemical interaction that causes the dextran to adsorb onto the PS particle surface. While of interest, this mechanism is specific to diffusant-particle systems, and cannot be accounted for in a diffusion model such as the one presented here. A recent paper by Nunnery et al. [45] shows that the adsorption of polymers in pores is sensitive to both the polymer MW and the pore dimensions. In fact, Nunnery et al. [45] demonstrate that for a known pore size, adsorption could be used to evaluate the MW of the polymer. This suggests that control of the pore size in the colloidal shell (through the diameter of the particles) may still be used to control adsorption and transport of large MW molecules.

4. Conclusions

Previous research suggests that colloidosome shells enable size-selective transport due to the particle-determined size of the pores between the surface particles [23,25,26]. They suggest that the presence of a colloidal shell reduces the rate of transport, and that larger molecules may be inhibited of diffusion through a closely packed colloidal shell.

In this paper we systematically investigate the effect of the dimensions of the colloidal particles composing the shell, which determine the surface porosity, and the size of the diffusing molecule on transport. Contrary to expectations, we find that the transport rate of molecules through the colloidal shell is insensitive to the size of the particles (unless the particle radius is of the same order of magnitude as the diffusing molecule). Coatings composed of 20 nm particles provide the same rate of release as that of 1 µm. Even increasing the particle size to 3.3 µm does not affect the release. These results are summarized in Eq. (4), which relates the size of the diffusing molecule to the volume fraction of the particles in the colloidal shell and the particle size. It should be noted, however, that Eq. (4) does not apply in systems where the diffusant molecule interacts with the colloidal particles through some chemical forces (such as dextran adsorbing on the PS particles in our studies).

Our results are in good agreement with previous studies [25,26]; Indeed, colloidal shells may used to suppress transport of larger molecules. However, we show that the shell-induced reduction in transport does not require close-packing of the particles. Also, we provide a more quantitative measure for the relationship between the properties of the colloidal shell (particle size, packing density) and the rate of transport.

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