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Research paper

Two galactomannans and scleroglucan as matrices for drug delivery: Preparation and release studies

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Abstract

Two galactomannans, Guar gum and Locust bean gum, have been used as matrices for tablets to study the release of model molecules. As a comparison, matrices obtained with another polysaccharide, Scleroglucan, have been tested. Despite the different conformations that the polymers assume in aqueous solution (flexible coils for Guar gum and Locust bean gum; triple helix for Scleroglucan), when prepared as tablets, they show (in distilled water and at 37 °C) very similar release profiles of guest molecules (i.e. theophylline, vitamin B12 and myoglobin) of different steric hindrance. Furthermore, the polymers were chemically crosslinked with glutaraldehyde to obtain a network suitable as a matrix for modified drug release. The delivery of the model molecules from the Guar gum and Locust bean gum gels, and from tablets prepared from the freeze-dried hydrogels of the three polymers was evaluated, and a comparison with the tablets prepared with the not-crosslinked polymers was carried out. Experimental data showed how the presence in the matrix of a well-defined network, by introducing a spacer among the macromolecular chains, always increased the rate of delivery of the tested molecules in comparison to the release profiles obtained when no crosslinker was present. Release data from the tablets were analyzed according to a mathematical model able to determine the relative importance of drug dissolution and drug diffusion on the overall release kinetics. Good agreement was found between the simulated and the experimental data.

Keywords: Galactomannans; Guar gum; Locust bean gum; Scleroglucan; Glutaraldehyde; Theophylline; Vitamin B12; Myoglobin; Modified release

1. Introduction

Galactomannans are neutral polysaccharides that occur in the endosperm of the seeds of leguminous plants. They consist of a β-(1 → 4)-D-mannose (M) backbone to which galactose (G) units are attached α-(1 → 6). The various galactomannans show a different mannose/galactose ratio, a different substitution pattern of side-chain units and different molecular weights [1–5]. In the present work two galactomannans, Guar gum (GG) and Locust bean gum (LBG) (Fig. 1b and c), have been considered for their possible use as matrices for modified drug delivery. Furthermore, another neutral polysaccharide, Scleroglucan [6–8] (Sclg) (Fig. 1a), has been tested and chosen for a comparison.

The primary structure of this last polymer exhibits a backbone built up by β-(1 → 3) linked β-D-glucopyranose (glcp) units with single glcp side chains linked β-(1 → 6) to every third residue in the main chain. GG has a mannose/galactose ratio (M/G) of about 2 while LBG has a M/G ratio of about 4; in aqueous solutions both polysaccharides assume a flexible coil conformation as it can be evidenced, for example, by the value of the Mark–Houwink–Sakurada exponent $\alpha = 0.74 \pm 0.01$ [9,10]. On the other side Sclg, in aqueous solution, assumes a very stable triple helix

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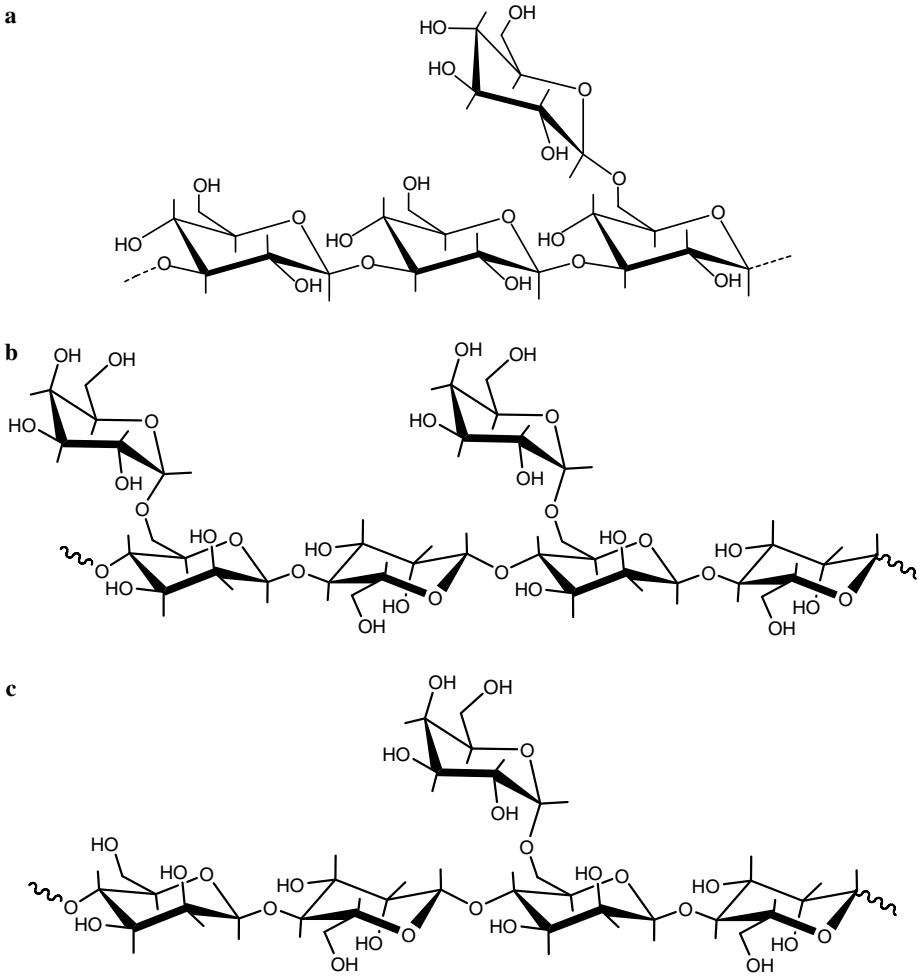

Fig. 1. Repeating units of scleroglucan (a), Guar gum (b) and Locust bean gum (c).

conformation and, with a value of $\alpha = 1.7$ [11], it represents one of the most stiff systems present in nature; accordingly, Sclg exhibits a rigid rod-behaviour in a wide range of pH and temperatures.

Furthermore, Sclg was extensively studied for its peculiar properties, notably rheological properties [12], and has been also proposed for biomedical applications [13–17]; and it is found, as viscousing agent, in some preparations for topical uses that are actually in the market.

Scleroglucan is also used in the oil industry, for food quality improvements, and in cosmetic formulations [18–20].

In the present work, the three polymers have been used as carriers for the delivery of three model molecules with different size, specifically theophylline (TPH), vitamin B12 (Vit. B12) and myoglobin (MGB). The release profiles in water from tablets prepared with GG, LBG and Sclg, loaded with the guest molecules, were evaluated. Crosslinked systems, obtained by crosslinking reactions of the mentioned polysaccharides with glutaraldehyde (Ga), were also studied (GG/Ga, LBG/Ga, and Sclg/Ga) [21,22].

According to the chemical structure of the repeating units of GG and LBG and to the fact that the reaction of Ga occurs only with the vicinal diols, the network formed in the presence of GG will be more crosslinked than that obtained with LBG due to the fact that the degree of branching is higher in the former polymer. No direct comparison can be made for the Sclg that, though bearing vicinal diols, has a different repeating unit and in particular a different conformational state.

In fact, also from the macroscopic point of view, the macromolecular networks gave gels capable to maintain their shape in a ''test-tube inverting method'' (i.e., self-sustaining gels) [23] in the case of GG and LBG while, in the case of Sclg, only an increase in viscosity leading to a connectivity not extended to the whole sample (i.e., microgels) was obtained. For this reason the releases from the gels of the model molecules were studied only for the GG and LBG systems while the release of TPH and MGB was monitored for all the three crosslinked polymers after the preparation of tablets.

It is very important to study the behaviour of these systems as it is well-known that the polysaccharidic matrices can play an important role in the design of drug release formulations since the bioavailability of many drugs is deeply

dependent on the performance of the carrier system [24–26].

It is therefore, very useful to know how, and to which extent, such type of matrices can be capable to modulate the release of guest molecules; in particular, in the present work, a comparison among the behaviours of matrices prepared with the plain polymers and those prepared after the crosslinking reaction is reported. Furthermore, the influence of the dimension of the model molecule on the release profiles has been evaluated; the influence of the different hydration, between gels and tablets, has also been considered.

Finally, release data have been studied by means of a recently developed semi-empirical mathematical model [27] aimed to yield some insight about the relative importance of the phenomena ruling release kinetics. In particular, this model assumes that tablet composition, drug dissolution and diffusion through the swelled polymer layer surrounding the tablet dry core, are key factors for the delivery process.

2. Materials and methods

2.1. Materials

All polymers, Sclg, GG and LBG, were purchased from Carbomer (U.S.A.). Sclg was provided with a degree of polymerization DP = 800 (MW $\sim 1.5 \times 10^6$); GG had a molecular weight, MW $\sim 1.2 \times 10^6$; and LBG a MW $\sim 1.8 \times 10^6$.

TPH and Ga were Carlo Erba products (Italy), Vit. B12 and MGB were purchased from Fluka (Germany). All other products and reagents were of analytical grade. For the preparation of the samples distilled water was always used.

2.2. Polymer purification

A given amount of polymer (GG, LBG or Sclg) was dissolved in distilled water, and then kept under magnetic and mechanical stirring for 24 h. The obtained solution was exhaustively dialyzed at 7 °C against distilled water with dialysis membranes of a cut-off 12,000–14,000, and then freeze-dried. The lyophilized product was stored in a desiccator until use.

2.3. Hydrogel preparation

Starting from literature data [22] the reaction conditions to prepare the crosslinked systems were optimized. A given amount of GG, LBG or Sclg (100 mg for the release experiments from the gels and 180 mg for the formation of the gels to be used for the preparation of tablets) was kept under magnetic stirring in distilled water for 24 h and at the appropriate concentration ($c_p = 1.5\%$ (w/v) for GG and LBG; $c_p = 0.5$ and 1.0 for Sclg) to obtain a complete dissolution of the polymer. A few drops of concentrated sulphuric acid were then added (to protonate the hydroxylic groups of the polymer) to the solution that was kept under magnetic stirring for 30 min. An appropriate amount of Ga was added to obtain a suitable value of r, where r represents the ratio between the crosslinker moles and the moles of repeating units of the polymer: $r =$ (crosslinker moles)/(moles of repeating units of polymer).

The composition of all the samples tested in the present study is summarized in Table 1.

For GG and LBG the formulations reported in Table 1 ($c_p = 1.5\%$ and $r = 4.0$) were chosen among the numerous compositions that were tested and they correspond to the minimum value of c_p that gave self-sustaining gels.

The reaction mixtures were stirred for another 30 min and kept in a thermostatted bath at 30 °C for 48 additional hours for gel setting. The samples were dialyzed, using dialysis membranes with a cut-off 12,000–14,000, at 7 °C until the unreacted Ga disappeared from the dialysis solvent. The presence of polymeric Ga was detected at 235 nm and the monomeric Ga at 280 nm. In fact it is well-known that Ga exists in solution in various forms [28–30] and therefore it is not possible to give an exact description of the resulting network. Aqueous solutions of the aldehyde consists actually of a mixture of free aldehyde, mono- and dihydrated monomeric glutaraldehyde, monomeric and polymeric cyclic hemiacetals and various α,β-unsaturated polymers; nevertheless Ga has been used as a crosslinking agent more frequently than any other reagent since it is less expensive, it is readily available and it is highly soluble in aqueous solution.

The obtained self-sustaining gels, settled in a 10-ml beaker, showed the geometry of a cylinder having a diameter of 22 mm and a height of 18 mm. Unfortunately, such good conditions did not occur for Sclg, although different test samples were prepared and a partial list of them is given in Table 1.

The hydrogels were then used to study the release of TPH, Vit. B12 and MGB or lyophilized for the preparation of tablets. A different procedure was performed to load the hydrogels with the different molecules (see below).

As above pointed out, self-sustaining gels were obtained only with GG and LBG, while in the case of Sclg microgel formation was observed; consequently, with this last

Table 1
List of the hydrogels prepared with Sclg, GG and LBG together with the polymer concentration c_p and r values

Polymer	c_p (%, w/v)	r
Sclg	0.5	0.5
Sclg	0.5	5.0
Sclg	1.0	0.5
Sclg	1.0	5.0
GG	1.5	4.0
LBG	1.5	4.0

For the last two polysaccharides the parameters represent the minimum value requested to obtain a self-sustained gel ($r =$ (crosslinker moles)/(moles of repeating units of polymer)).

Table 2
List of the samples, gels and tablets, together with the corresponding loaded model drug, studied in the present work

Polymer	Gel		Tablets		Tablets reference		
	TPH	MGB	TPH	MGB	TPH	Vit. B12	MGB
Sclg	–	–	X	X	X	X	X
GG	X	X	X	X	X	X	X
LBG	X	X	X	X	X	X	X

polymer, only release studies from tablets could be performed (see Table 2).

2.4. Hydrogel loading, release experiments from gels, preparation of tablets and dissolution experiments from tablets

2.4.1. GG and LBG

The dialysed hydrogels (obtained starting from ca. 100 mg of polymer) were freeze-dried and then soaked in a saturated solution of TPH for 48 h. The samples were then washed with distilled water to remove the excess of imbibition solution and then tested for the release of TPH. In the case of MGB, on the contrary, drug loading was carried out by directly inserting with a needle a given amount of an aqueous MGB saturated solution in the freeze-dried samples until the matrices were completely soaked up.

Release experiments were carried out by immersion of the gel in distilled water ($V_r = 200\ cm^3$; $T = 37\ °C$) at a fixed distance from the vessel bottom by means of a thin-web hosting the gels. Release medium was gently magnetically stirred. Three millilitre samples were withdrawn from the solution at appropriate time intervals and replaced with the same amount of fresh solvent. TPH and MGB concentrations were detected, respectively, at 272 and 409 nm by means of a spectrophotometer (Perkin-Elmer, lambda 3a, UV–vis spectrometer) equipped by 1.0 cm path-length quartz cells.

Tablets were prepared by compressing with an IR die (Perkin-Elmer hydraulic press; compression force = 5.0 kN) the freeze-dried hydrogel (obtained starting from ca. 180 mg of polymer) containing the model drug.

Reference tablets, i.e., those tablets made up of not-crosslinked polymer, were prepared by freeze-drying aqueous solutions containing the polymer and the model drug (see Table 3) followed by compression at the same conditions above reported to obtain the final dosage forms.

Table 3
Molecular weight (MW) of model molecules and their radius of van der Waals (r.v.Waals)

Model molecule	MW	r.v.Waals (Å)
TPH	198.18	3.7
Vit. B12	1355	8.5
MGB	17,800	21.0

All tablets had a weight of 190 ± 10 mg, a diameter of 13.00 ± 0.05 mm, and a thickness of 1.50 ± 0.10 mm.

Dissolution experiments from tablets were carried out according to USP XXIV, using the rotating basket apparatus at 37.0 ± 0.1 °C, 100 rpm and distilled water (500 ml, pH 5.4) as a dissolution medium. Aliquots of the dissolution medium (5 ml) were taken at fixed time intervals, the same amount of fresh solvent was added in the apparatus and TPH, Vit. B12 (in the case of the reference tablets) or MGB concentration was spectrophotometrically measured at 272, 361 and 409 nm, respectively. Both, release and dissolution experiments, were carried out in triplicate. The values reported in the present paper represent mean values and lay within 10% of the mean.

2.4.2. Sclg

As mentioned before, in this case, although different c_p and r values were considered for the crosslinking reaction, it was impossible to obtain self-sustaining gels. Accordingly, only tablet systems were considered. Tablets were prepared from reaction mixtures containing the appropriate amount of polymer and crosslinker (see Table 1) followed by dialysis, lyophilization and immersion in distilled water, where model drugs were previously solubilized. Finally, these systems underwent a further freeze-drying and then a compression process (Perkin-Elmer hydraulic press; compression force = 5.0 kN) to obtain the final drug-loaded tablets.

Reference tablet preparation followed the same method with the only difference that the crosslinker agent was not added.

All tablets had a weight of 200 ± 10 mg, a diameter of 13.00 ± 0.05 mm and a thickness of 1.50 ± 0.10 mm.

Dissolution experiments from tablets were carried out in triplicate, according to the method above described for GG and LBG.

The values reported in the present paper represent mean values and lay within 10% of the mean.

2.5. Water uptake

Water uptake was evaluated, in distilled water at 37 °C, by the relative increase of weight of tablets prepared with GG and tablets obtained starting from the freeze-dried hydrogel GG/Ga. The experiments were carried out in triplicate.

The values reported in the present paper represent mean values and lay within 10% of the mean.

2.6. Mathematical modelling: release from tablets

Drug-release kinetics from tablets prepared with hydrophilic polymers is affected by many factors such as polymer swelling, polymer erosion and drug-dissolution characteristics, beside drug/polymer ratio and tablet geometric features [31,32].

Although many powerful mathematical models were developed on this topic [33], very often they are not so user-friendly as they require numerical solutions. Accordingly, in this work, experimental data will be analyzed by means of a recent, more user-friendly, semi-empirical model [27].

Briefly, the tablet is seen as a uniform dispersion of two non-interacting phases where only one phase (drug) can dissolve, while the second one (polymer) can swell and can undergo erosion. As a consequence, drug release will be affected by two resistances identifying the drug dissolution and the diffusion through the swelled layer surrounding the dry tablet core (the swelled layer is delimited by the swelling front, on its inner side, while it is delimited by the erosion front on its outer side). Accordingly, remembering that the global resistance to diffusion exerted by a multi-layered membrane is given by the sum of the resistance of each layer [34] and that the inverse of the intrinsic dissolution constant k_d can be thought as the drug dissolution resistance, the model differential form reads:

$$\frac{dC_r}{dt} = -\frac{x_d A}{V_r}\frac{C_s}{\left(\frac{1}{fk_d} + R\right)} \tag{1}$$

where C_r and C_s represent, respectively, drug concentration and solubility in the receiver environment, t is time, A is the time-dependent release surface area at the diffusion front, x_d is the drug mass fraction at the swelling front (it accounts for the fact that drug release depends on the effective drug-dissolution area at the diffusion front that can be approximated by the product Ax_d), V_r is the receiver volume and f is a parameter accounting for the fact that, due to the presence of the swelled layer, drug-dissolution constant k_d will be lower than that relative to the pure solid drug dissolution [35].

Finally, R, indicating the resistance to diffusion offered by the swelled layer, can be assumed directly proportional to the swelled layer thickness [27]. On the basis of experimental data, R time-dependence can be of different types, according to the following equations [36]:

$$R(t) = R_e^0(1 - e^{-k_{re}t}) \tag{2}$$

$$R(t) = R_e^0\, e^{-k_{re}t} \tag{3}$$

where R_e^0 and k_{re} are two model fitting parameters. According to Eq. (2), swelled layer formation is supposed to be slower than drug dissolution so that, at the beginning (time zero), swelled layer resistance is zero (this corresponds to an infinite permeability value of swelled layer being its thickness vanishing) and it increases with time until approaching an asymptotic value (R_e^0). On the contrary, Eq. (3) accounts for a very fast (ideally, instantaneous) swelled layer build-up in comparison to drug dissolution so that its thickness reaches a maximum value (R_e^0) at the beginning and, then, it decreases due to erosion. In order to get a model analytical solution it is now necessary to define the A-dependence on C_r. Assuming, for the sake of simplicity, that the ratio K between the diffusion front height h and the radius R_a is always constant during the release process, A can be estimated according to [27]:

$$A = (\alpha - \beta C_r(t))^{2/3} \tag{4}$$

$$\alpha = \frac{(2\pi(1+K))^{3/2} M_0}{\pi K C_0} \quad \beta = \frac{(2\pi(1+K))^{3/2} V_r}{\pi K C_0} \tag{4'}$$

where M_0 and C_0 are, respectively, the initial drug amount and concentration contained in the tablet. Substituting Eq. (4) into Eq. (1), we finally get:

$$C_r^+ = \frac{C_r}{M_0/V_r} = 1 - \left(1 - \frac{2(1+K)x_d}{3M_0^{1/3}}\left(\frac{\pi}{K^2 C_0^2}\right)^{1/3} F(t)\right)^3 \tag{5}$$

where $F(t)$ is given by:

$$F(t) = C_s\left[\frac{t}{R_e^0 + 1/fk_d} + \frac{\ln\,(1 + R_e^0(1 - e^{-bt})fk_d)}{(R_e^0 + 1/fk_d)k_{re}}\right] \tag{6}$$

if Eq. (2) holds and

$$F(t) = fk_d C_s\left[t + \ln\left(\frac{R_e^0\, e^{-bt} + 1/(k_d f)}{B + 1/fk_d}\right)\Big/ k_{re}\right] \tag{7}$$

if Eq. (3) holds.

In order to take into account the fact that an incomplete release from tablets can take place, Eq. (5) can be multiplied for a constant, named R_{id} and ranging between 1 and 0, that can be simply estimated on the basis of the drug amount released after a very long time.

3. Results and discussion

3.1. Water uptake experiments

In Fig. 2a water uptake of tablets prepared with only GG, LBG and with GG/Ga and LBG/Ga is reported. It is evident how the matrix prepared with only GG showed a lower swelling capacity while, in the case of the tablets prepared from the hydrogel GG/Ga, a remarkably higher amount of water was uptaken. The same data, reported as a function of the square root of time (Fig. 2b), indicated that the phenomenon, at least macroscopically, could be satisfactory described according to a Fickian process.

The significative difference in the amount of water in the GG/Ga tablets during the swelling process represents one of the parameters that affect the release of the model drugs (see below). On the opposite, in the case of the LBG systems, the difference between the matrices with and without

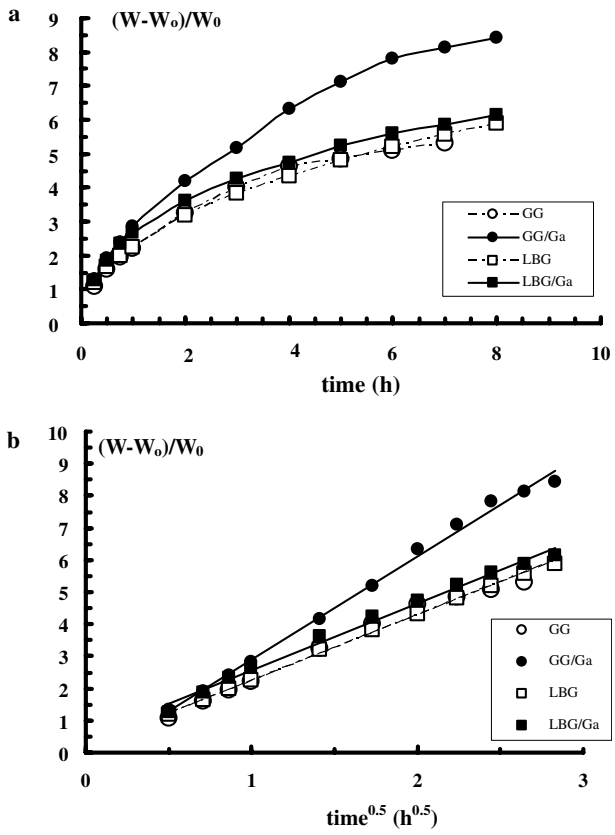

Fig. 2. Water uptake from tablets of GG, LBG (empty symbols), GG/Ga and LBG/Ga (full symbols). (a) As a function of time; (b) as a function of the square root of time. Experiments were carried out in triplicate and the obtained values always laid within 10% of the mean.

Ga is much less remarkable though a slightly higher value is always found for the LBG/Ga tablets. Also for these tablets the water penetration followed a simple diffusion process, as evidenced in Fig. 2b.

3.2. Release experiments from gels

The release experiments from the gels were performed, as above explained, only with samples of GG/Ga and LBG/Ga (see Table 2).

In Fig. 3 the release profiles are reported as $(M_t/M_\infty) \times 100$ where M_t represents the amount of model drug found in the release medium at time t and M_∞ represents the amount of model drug released after an infinite time, i.e., the total amount of the drug initially present in the hydrogel.

In Fig. 3 it is possible to observe the relevant difference between the releases of TPH and MGB. The first molecule is completely delivered within the first 8-h while MGB shows an initial slight release (almost 20%) already after 15 min, but no further increase can be detected for the following duration of the experiment (24 h). This behaviour can be explained in terms of the different steric hindrance of the two molecules in comparison with the mesh size of the polymeric network: TPH, with a radius

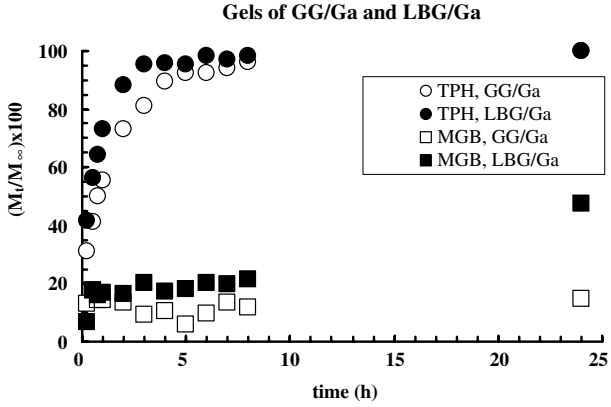

Fig. 3. Release profiles, in distilled water at 37 °C, of TPH and MGB, from gels of GG/Ga and LBG/Ga with $c_p = 1.5\%$ (w/v) and $r = 4$. The solid line is only a guide to the eye. Experiments were carried out in triplicate and the obtained values always laid within 10% of the mean.

of 3.7 Å (Table 3), is freely released from the gel and it can totally diffuse out of the matrix while MGB, with a radius of 21.0 Å (Table 3), is not capable to move through the meshes. This reflects into a two-step MGB release behaviour as about 20% of the initial dose, present in the most outer layers, can be delivered in the surrounding medium since the beginning of the experiment; on the other side, the remaining 80% behaves as if it was permanently entrapped within the hydrogel network, at least within the time of the experiment (24 h), as already found with another polysaccharidic matrix [37].

Furthermore, with the LBG polymer, as above pointed out, the number of crosslinks formed with Ga is lower than those present in the GG/Ga system and this implies the formation of wider meshes. According to this fact, the TPH experiments with a looser network lead to a faster diffusion of the guest molecules through the hydrogel. This explains why the release of TPH from the LBG/Ga gel is faster than that from the GG/Ga gel and only 3 h are required for a complete delivery of the molecule. Similarly, a slight increase of MGB release is observed for LBG/Ga gels with respect to that obtained with GG/Ga.

3.3. Release experiments from tablets

In Fig. 4 the release profiles of TPH, Vit. B12 and MGB from tablets prepared with the polysaccharides without crosslinker are reported.

First of all it can be observed that the delivery curves, as expected, follow a trend related to the molecular dimensions of the molecules (see Table 3): the release rate of TPH is the highest one and almost the total amount of the drug is found in the dissolution medium after 24 h, the delivery rate of Vit. B12 is lower and not all the vitamin loaded in the matrix is released within 24 h, the delivery rate of MGB is very low and after 24 h only a reduced amount of the loaded molecule is delivered. Furthermore, as far as the data of the first 8-h are concerned, no

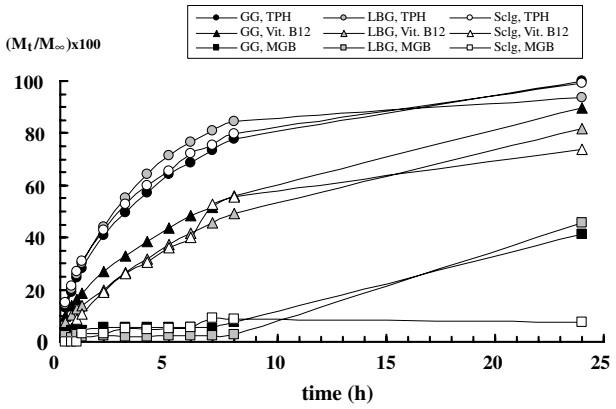

Fig. 4. Release profiles, in distilled water at 37 °C, of TPH, Vit. B12 and MGB, from tablets prepared with the GG, LBG and Sclg polymers. The dotted lines are only a guide to the eye. Experiments were carried out in triplicate and the obtained values always laid within 10% of the mean.

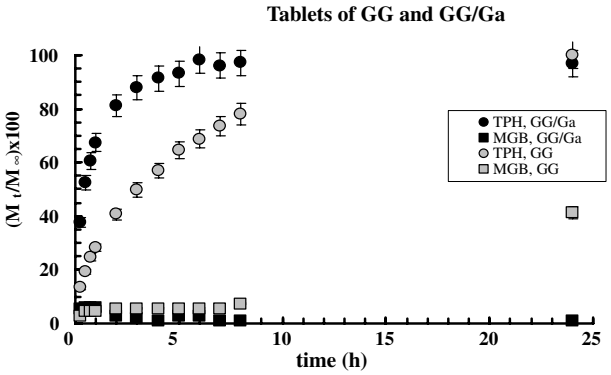

Fig. 5. Release profiles of TPH and MGB, in distilled water at 37 °C, from tablets of GG prepared with and without Ga. Experiments were carried out in triplicate and the obtained values always laid within 10% of the mean.

significant differences can be detected among the three polymers with respect to the release of each guest molecule. This result appears to be quite important because it proves that, regardless of the different chemical structure and conformational characteristics of the three polysaccharides, their release behaviour is similar. Nevertheless some differences can be observed after 24 h: in the case of the smallest molecule, TPH, the three release curves do not vary significantly while a slight difference can be appreciated in the case of Vit. B12 where the lowest amount released refers to the Sclg tablet. Finally, for MGB, an evident difference can be observed between the Sclg tablets on one side and the GG and LBG tablets on the other side: in the case of Sclg even at 24 h the release is very low, about 10%, while in the case of GG and LBG tablets a 40% of MGB is actually delivered after the same time interval. Such difference can be ascribed to the different microenvironment experienced by the MGB, being GG and LBG both flexible polymers while Sclg is one of the most rigid polysaccharides. After the imbibition step, the viscosity of the swelled tablets can be lower in the case of the more flexible chains and much higher for the rigid Sclg, as it happens in dilute solutions of these polymers.

In Fig. 5 the releases of TPH and MGB from the tablets prepared with GG, with and without Ga, are compared. It is evident how the delivery is again strongly dependent on the molecular dimensions of the guest molecule. The introduction of crosslinking offers more space to the diffusion out of the matrix for TPH; on the other side if such statement can be reasonable for a small molecule like TPH, in the case of a larger molecule like MGB an opposite effect takes place because the large molecule is somehow entrapped within the matrix, at least for the first 8-h. Only after 24 h there is an appreciable delivery of MGB (about 40%) from the system without Ga which can anyhow be related to a partial erosion of the matrix.

As observed from water uptake experiments (see Fig. 2) the GG tablets absorb less water than the GG/Ga tablets and this is another parameter that influences, together with the presence of the crosslinking, the faster delivery of a molecule like TPH. On the other side, in the case of a larger molecule, like MGB, the effect of a higher water uptake is practically overtaken by the presence of the meshes that do not allow the delivery of the molecule that is released only because of a partial erosion of the matrix.

In Fig. 6 the release of TPH and MGB from the tablets prepared with LBG crosslinked with Ga and the release from the tablets prepared with the polymer without the crosslinker are compared. Some aspects are quite relevant: the release of the model molecule, in the case of GG (Fig. 5), is faster when the Ga is present while no differences are evident in the case of LBG. This behaviour can be explained in the following manner: if we consider that LBG has only a few side chains in comparison to the GG, only a reduced number of meshes can be developed within the network that cannot significantly influence the mechanism of the release of TPH. Even if the amount of Ga is stoichiometrically in excess ($r = 4$), not all possible effective linkages (i.e., those between two polymeric chains) will be formed and therefore, although a self-sustaining gel

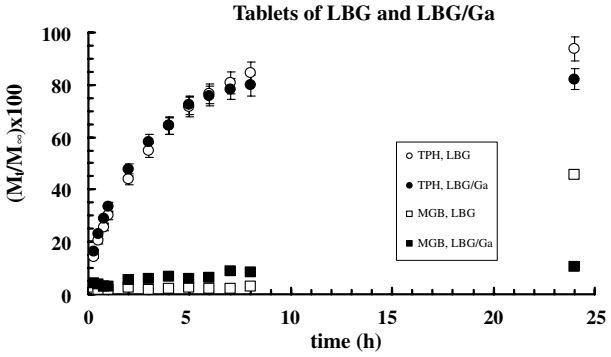

Fig. 6. Release profiles of TPH and MGB, in distilled water at 37 °C, from tablets of LBG prepared with and without Ga. Experiments were carried out in triplicate and the obtained values always laid within 10% of the mean.

is obtained, only a few inter-chain chemical connections are actually formed.

On the other side, in the case of GG/Ga (Fig. 5), the systems have a high crosslinking degree, due to the higher number of side chains present in this polysaccharide, that influences macroscopically the delivery of a small guest molecule such as TPH.

The proposed explanation of the different trends of TPH release from GG/Ga and LBG/Ga can be further supported by the empirical observation that, although both gels are self-sustaining, LBG/Ga hydrogels appear to be remarkably weaker than the corresponding GG/Ga hydrogels.

In Fig. 7 the release of TPH and MGB from tablets prepared with Sclg with different amounts of Ga is reported.

Again, as expected according to the data relative to GG, the slowest release rate of TPH is found for the system without crosslinker. Introducing a small amount of Ga ($r = 0.5$) a faster release of TPH is observed and the same behaviour is found even with an excess of crosslinker. Obtained results indicate how it is sufficient to introduce a small amount of Ga ($r = 0.5$) to form spaces in the entangled system that improves noticeably the release of the guest molecule. By addition of an excess of Ga ($r = 5.0$) the same release profile is obtained, thus it can be asserted that the basic structure of the network obtained for $r = 0.5$ does not change when $r = 5.0$ and the diffusion of a small molecule like TPH is consequently not affected.

Again, as in the cases previously discussed, the release of MGB is almost negligible because the large molecule is entrapped within the matrix.

The results referring to data analysis (release from tablets), according to the mathematical model above reported, are given in Table 4. Model best fitting parameters are determined knowing that drug mass fraction at the swelling front x_d is equal to 0.1, tablet height/radius ratio K is equal to 0.215 and that the initial drug amount M_0 and concentration C_0 in the tablet are, respectively, 20 mg and 107.6 mg/cm^3. Additionally, while TPH solubility (37 °C) is equal to 12.5 (mg/cm^3), Vit. B12 and MGB solubility

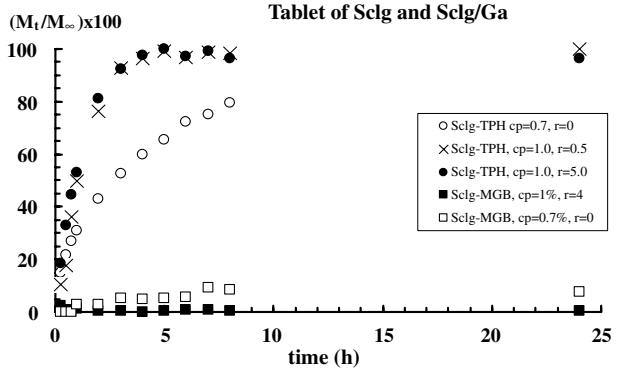

Fig. 7. Release profiles of TPH and MGB, in distilled water at 37 °C, from tablets prepared with only Sclg and Sclg/Ga at different c_p and r values. Experiments were carried out in triplicate and the obtained values always laid within 10% of the mean.

Table 4
Model best fitting parameters (R_e^0, k_{re}, f) and F statistic value referring to TPH and Vit. B12 release from tablets

	F	R_e^0 (h/cm)	k_{re} (1/h)	(1/(fk_d)) (h/cm)	R_{id}
TPH					
GG	64137	1.34	0.92	0.25	1.00
LBG	3358	1.06	0.51	0.33	0.93
Sclg	61813	1.38	0.94	0.20	1.00
GG/Ga	3798	34.1	0.01	0.09	1.00
LBG/Ga	1482	1.53	0.21	0.28	0.84
Sclg/Ga	3560	664	10^{-4}	0.24	1.00
Vit. B12					
GG	37589	3.61	1.02	0.41	0.92
LBG	35264	3.51	2.50	0.21	0.83
Sclg	1204	1.48	12.65	1.00	0.74

Fitting is performed knowing that $k_d = 11.28$ cm/h for TPH, while for Vit. B12 a very large k_d value has been assumed (10^3 cm/h). Swelled layer resistance is given, for both molecules, by Eq. (2).

(37 °C) are equal to 19.7 and 0.93 mg/cm^3, respectively. Due to the practical impossibility of discerning the drug-dissolution constant k_d from the fitting parameter f on the basis of our data, we prefer to consider the whole (f k_d) product as a unique model fitting parameter. This parameter accounts for drug dissolution inside the polymeric matrix. It will be clear in the following part that this choice does not substantially affect what we can infer from our model data fitting. The parameter R_{id}, accounting for a possible not complete release from the tablet, is not considered as a real fitting parameter as it is evaluated on the basis of the drug amount released after a very long time (24 h in this study). Accordingly, R_e^0, k_{re} and fk_d are model fitting parameters.

In the TPH case, Table 4 shows that model best fitting is always good regardless of the fact we are dealing with not crosslinked (GG, LBG, Sclg) or with crosslinked polymers (GG/Ga, LBG/Ga, Sclg/Ga) (we remember that the model is satisfactory if the fitting associated F statistic value, reported in Table 4, is higher than the theoretical one that, in this case, reads $F(2, 9, 0.99) = 8.02$) [38].

Interestingly, only the assumption that gel resistance develops according to Eq. (2) (consequently, Eq. (6) applies in Eq. (5)) leads to a good data fitting. This means that, regardless of the kind of polymer, at the beginning, drug release is mainly ruled by drug dissolution and only later drug diffusion through the swelled layer plays a relevant role. Additionally, Fig. 8 clearly shows how, in the presence of crosslinking, the system resistance R is lower than that corresponding to not crosslinked systems for the first 5-h in the GG case, for the first 6-h in the LBG case and for the first 12-h in the Sclg case. This absolutely agrees with the experimental evidences about faster observed release kinetics from all crosslinked systems and, additionally, this is also in line with crosslinked systems faster swelling behaviour (see Fig. 2).

Also in the case of Vit. B12 case, model best fitting is always satisfactory regardless of the type of polymer (see

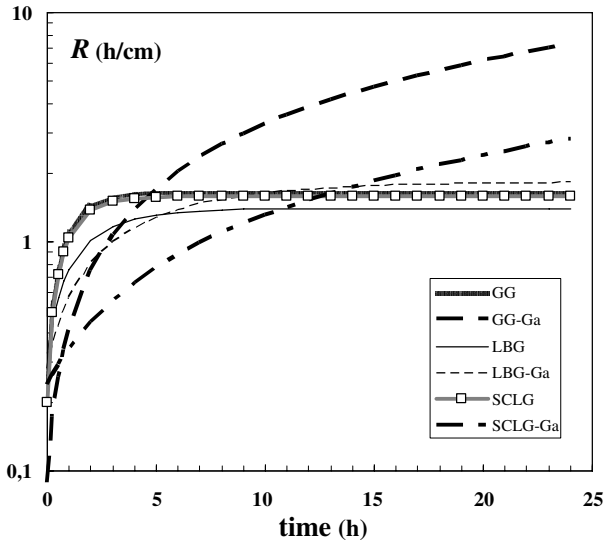

Fig. 8. Calculated resistance time-dependence R [Eq. (2)] for the various tested tablets.

F statistic values in Table 4). Again, as data fitting requires that swelled layer resistance (R) evolution be described by Eq. (2), and consequently, Eq. (6) applies to Eq. (5), Vit. B12 release is initially ruled by dissolution and then by diffusion through the swelled layer. Nevertheless, now, the effect of swelled layer presence is much more important than in the case of TPH due to the larger dimensions of Vit. B12 molecule. Indeed, model best fitting parameters (namely, R_e^0, k_{re} and fk_d) make it clear that R evolution is faster for Vit. B12 and that it reaches bigger final values than those obtained with TPH (higher k_{re} and R_e^0 values, see Table 4). The fact that R_{id} for Vit. B12 (fraction of drug released after 24 h) is, on average, smaller than for TPH could be explained observing that the bigger is the model molecule, the higher is the possibility to be trapped inside the polymeric network.

Finally, in the case of MGB, data fitting is not very significant due to the very small release kinetics observed. Nevertheless, the model would suggest that swelled layer formation is ruled by Eq. (3) (accordingly Eq. (7) applies in Eq. (5)), this meaning an instantaneous swelled layer formation followed by erosion. Consequently, MGB dissolution would never exert an important role and only drug diffusion through the swelled layer would be important. This frame seems to be compatible with the big dimensions of MGB molecule.

4. Conclusions

From the experimental data it can be concluded that the GG and Sclg polymers, when crosslinked with Ga and used for the preparation of tablets, increase the rate of release of small guest molecules because the chemical reaction with Ga introduces meshes with a size larger than those present in the simply entangled systems. Such effect is almost undetectable in the case of LBG where the degree of crosslinking is remarkably lower in comparison to the other two polymers. Furthermore, the GG tablets evidenced a lower water uptake than the GG/Ga tablets, this being another parameter that leads to a faster release from the networks. Thus, it can be assumed that the crosslinker acts as a spacer forming a network with sufficiently large meshes that are capable to lead to an easier solvent penetration and consequently to a faster delivery with respect to the matrices prepared with the plain polymers; in this last case the tablets, in the presence of water, form an entangled network with very high local viscosity whose porosity is very limited. Such effect cannot be evidenced when the guest molecule has dimensions comparable to or bigger than the size of the meshes.

The effect of crosslinking is even better shown when the release experiments are carried out directly from the hydrogels (Fig. 3) i.e., from a swelled network. In comparison to a hypothetical formulation containing the polymer solution without crosslinking connections that should lead to an instant delivery, the self-sustaining structures slowed down the release of model drugs quite noticeably especially for the largest tested molecule (MGB).

The present work shows how it is possible to modulate the release of molecules of different size from chemical hydrogels prepared with GG or LBG and Ga as a crosslinker. The effect can be evidenced also when the networks are used to prepare tablets.

As far as Sclg is concerned, a slightly different comment has to be added: tablets prepared only with the polymer showed, at 24 h, a reduced release of medium size (Vit. B12) and large molecules (MGB), with respect to that obtained with GG and LBG, while, when crosslinked, the effect in the delivery is qualitatively similar to that obtained with the other two polysaccharides: the introduction of meshes in the network leads always to a faster diffusion of the small guest molecule.

On a molecular level, solute diffusion through swollen gels depends on the relative size of the diffusing species and the mesh formed by the macromolecular chains. Therefore, morphological features associated with reduced macromolecular chain mobility and barriers to solute diffusion are also associated with decreased solute diffusion coefficient. Further information about the relationship between structure and release kinetics will be acquired, in the near future, from rheological studies on the tested systems.

The proposed mathematical model reveals to be an interesting tool to provide some insight about the release kinetics process with particular attention to the relative importance of drug-dissolution resistance and the resistance exerted by the swelled layer surrounding the tablets.

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