

Grafted Glycopolymer-Based Receptor Mimics on Polymer Support for Selective Adhesion of Bacteria

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ABSTRACT A sugar-containing monomer (2-lactobionamidoethyl methacrylate, LAMA) was grafted on a polypropylene (PP) microfiltration membrane surface by UV-induced graft copolymerization. The degree of grafting can be controlled by variation of monomer concentration, UV irradiation time, and photoinitiator concentration. Fourier transform infrared spectroscopy and scanning electron microscopy were employed to confirm the surface modification on the membranes. The water contact angle was used to evaluate the hydrophilicity change of the membrane surface before and after modification. Bacteria capture experiments showed that the membrane could selectively bind *E. faecalis* while adhesion of *S. maltophilia* was not influenced by the functionalization of PP with grafted poly(LAMA). The adhesion of *E. faecalis* onto poly(LAMA) grafted membrane could be inhibited by 200 mM galactose solution; however, glucose solution showed no inhibition effect. Moreover, occupying sugar residues on the membrane surface primarily by a galactose targeting lectin, peanut agglutinin, could significantly suppress the following adhesion of *E. faecalis*. All these results clearly demonstrate that this poly(LAMA) grafted PP membrane can selectively capture *E. faecalis* and that this selection is based on the interaction between galactose side groups on grafted flexible functional polymer chains on the membrane surface and galactose binding protein on the *E. faecalis* cell membrane.

KEYWORDS: glycopolymer • polypropylene microfiltration membrane • surface modification • bacteria adhesion

INTRODUCTION

Microorganisms (viruses, bacteria, fungi, and parasites) cause infections and diseases due to colonization and reproduction in the human body (1). The first step of bacterial infection was shown to be the irreversible adhesion to the surface of human cells (2). Different models and theories have been established to describe interaction mechanisms between surfaces causing irreversible bacterial adhesion because of universal attractive forces such as Lifshitz-van der Waals, acid–base, and electrostatic interactions (3, 4). On the basis of these models, an important topic of current research is to develop antiadhesive strategies to prevent or control bacterial adhesion (5, 6). On the other hand, surfaces with selective recognition properties are of great interest, especially in medical devices or for sensors. With respect to bacterial infection of human cells, the carbohydrate–protein interaction between the respective cell surfaces is the most often perceived adhesion mechanism (7). The development of receptor mimics for rapid and sensitive diagnosis of infections mediated by microorganisms has created an opportunity to improve treatment of disease and their prevention.

Carbohydrate–protein interaction is one of the most important interactions in biological systems (8, 9). It relies on the highly specific recognition between hydroxyl groups on a carbohydrate and the carbohydrate binding domain in a protein. Proteins that bind to carbohydrates, such as carbohydrate-specific enzymes and antibodies, occur widely in nature. Another class of such proteins which are called lectins has no catalytic activity and is also not a product of immune response (10). This recognition is widely accepted to be the key in a variety of biological processes and the first step in numerous phenomena based on cell–cell interactions, such as blood coagulation, immune response, infection, inflammation, embryogenesis, and intercellular signal transfer (11, 12). However, it is also known that individual carbohydrate–protein interactions are weak, with binding affinities which range between 10^3 and 10^4 M⁻¹ (13). In nature, this problem is solved by aggregation of the sugar receptors displayed on the cell surface into higher-order multivalent structures. The strength of binding and also its specificity can be improved by multivalent interactions, which have been found quite regularly in biological systems (14, 15). A large number of different synthetic multivalent glycoconjugates (glycoclusters, glycodendrimers, glycopolymers, etc.) have been designed to interfere effectively in carbohydrate–protein interactions and to facilitate the investigation of the multiple interactions occurring during these recognition events (16–20). Some works reported various multivalent synthetic glycoconjugates which exhibit antibody-like selectivity for molecules on the cell surface of pathogenic microorganisms. However, most of these re-

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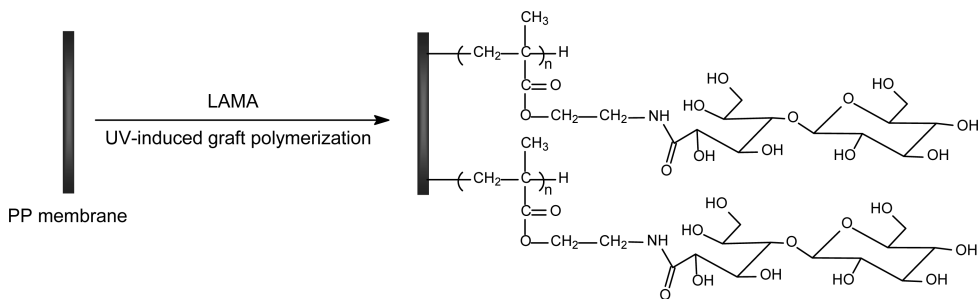
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Scheme 1. Schematic Representation for the UV-Induced Heterogeneous Graft Copolymerization of 2-Lactobionamidoethyl Methacrylate (LAMA) onto a Polypropylene (PP) Support Membrane Surface



search efforts were focused on viruses and toxins (21–23). In the latest publications, it has already been demonstrated that synthetic glycoconjugates can also have the ability to recognize exposed molecules on bacteria surfaces (24–26). Currently, we are working on immobilized receptor mimics for selective recognition of bacteria by surface-grafted linear glycopolymers on solid supports such as filtration membranes. This is highly attractive because this should provide the chance to combine high valency for binding, easy control of the molecular structure, and facile ways to vary the type of sugar ligands along with the advantages of immobilization on a solid support (27, 28).

In our previous work (29–32), glycopolymers were grafted onto a porous support, a polypropylene (PP) microfiltration membrane, to investigate sugar–protein interactions (Scheme 1). We had demonstrated that glycopolymers on the PP surface exhibit outstanding resistance toward non-specific protein adsorption and can effectively recognize lectins with high specificity. Analogous results have been obtained with glycopolymers grafted on sensor surfaces and analyses with surface plasmon resonance (33). In this work, we report the use of poly(2-lactobionamidoethyl methacrylate) (poly(LAMA)) to mimic the glyco-receptor on cell surfaces for specific recognition and binding of bacteria (cf. Scheme 1). LAMA is a glycomonomer bearing a cyclic galactose residue which can be recognized by *E. faecalis* (14, 34). This bacterial strain and *S. maltophilia*, which was taken as a negative control, were used to estimate the selective bacteria capture property of a poly(LAMA) functionalized membrane. With the glycopolymer on the surface, membranes are supposed to be promising porous adsorbers for specific bacteria capture which may be a very powerful tool in laboratory and industry.

EXPERIMENTAL SECTION

Materials. PP microfiltration membranes (type 2E HF) were purchased from Membrana GmbH, Germany; those had been prepared by a thermally induced phase separation method and had an average pore size of 0.20 μm and a relatively high porosity of about 80%. Before surface modification, the membranes were washed with acetone for 0.5 h to remove any impurities from the surfaces, dried in a vacuum oven in 40 $^{\circ}\text{C}$ for 1 h, and then stored in desiccators. LAMA was synthesized using a method described elsewhere (29, 34). Benzophenone (BP) and heptane were from Fluka and were used in p.a. grade without further purification. The water used in all syntheses and measurements was from a Milli-Q system. Peanut agglutinin

(PNA) and concanavalin A (Con A) were from Vector Laboratories, and BSA was from Sigma. All proteins were used as received. The bacterial strains were obtained from the German Collection of Microorganisms and Cell Cultures (DSMZ, Germany). *Enterococcus faecalis* (*E. faecalis*), DSM 20478, was maintained and cultivated on Corynebacterium agar at 36 $^{\circ}\text{C}$. *Stenotrophomonas maltophilia* (*S. maltophilia*) was maintained and cultivated on Standard 1 agar at 30 $^{\circ}\text{C}$. For all bacterial adhesion experiments, 10 mM *N*-2-hydroxyethylpiperazine-*N'*-2-ethanesulfonic acid (HEPES) buffer (pH 7.4) containing 0.1 mM Ca^{2+} was used.

Surface Modification of PP Membranes. Poly(LAMA) was grafted to the PP membrane surface (cf. Scheme 1), by a previously reported two step UV-induced graft copolymerization with PP “entrapped” BP as photoinitiator (35). Briefly, PP membrane was dipped into a 10 mL photoinitiator solution (benzophenone in heptane) for 60 min and then dried in air for 30 min. Thereafter, the membrane was washed with acetone and quickly wiped with filter paper. The membrane with pores still wetted by acetone was fixed between two filter papers immediately and immersed into a 10 mL monomer solution (LAMA in water) in a Petri dish. Then, UV irradiation was done for a predetermined time under argon gas environment. Finally, the membrane was washed with water intensively and dried in a vacuum oven at 40 $^{\circ}\text{C}$ to a constant weight. When grafting conditions (monomer concentration, UV-irradiation time, and photoinitiator concentration) are adjusted, the degree of grafting (DG) can be controlled, here defined as

$$\text{DG} = \frac{W_1 - W_0}{A}$$

where W_0 and W_1 are weight of unmodified and poly(LAMA) grafted membrane, respectively, and A is the outer surface area of the support membrane.

Surface Characterization. To investigate the surface chemical structure and morphology before and after the modification and to confirm the grafting, the following surface characterization techniques were used: attenuated total reflectance Fourier transform infrared spectroscopy (FT-IR/ATR), scanning electron microscopy (SEM), and water contact angle measurement.

FT-IR/ATR measurement was carried out on a Vector 22 FT-IR (Varian 3100, USA) equipped with ATR cell (KRS-5 crystal, 45 $^{\circ}$). Sixty-four scans were taken for each spectrum at a resolution of 4 cm^{-1} . SEM images were taken on a Field Emission SEM (SIRION, FEI, U.S.A.). An OCA20 contact angle system (Dataphysics, Germany) was used for the determination of air/water contact angles at room temperature. Static contact angle was measured by sessile drop method as follows. First, a water drop ($\sim 5 \mu\text{L}$) was lowered onto the membrane surface from a needle tip. Then, the images of the droplet were recorded. Contact angles were calculated from these images

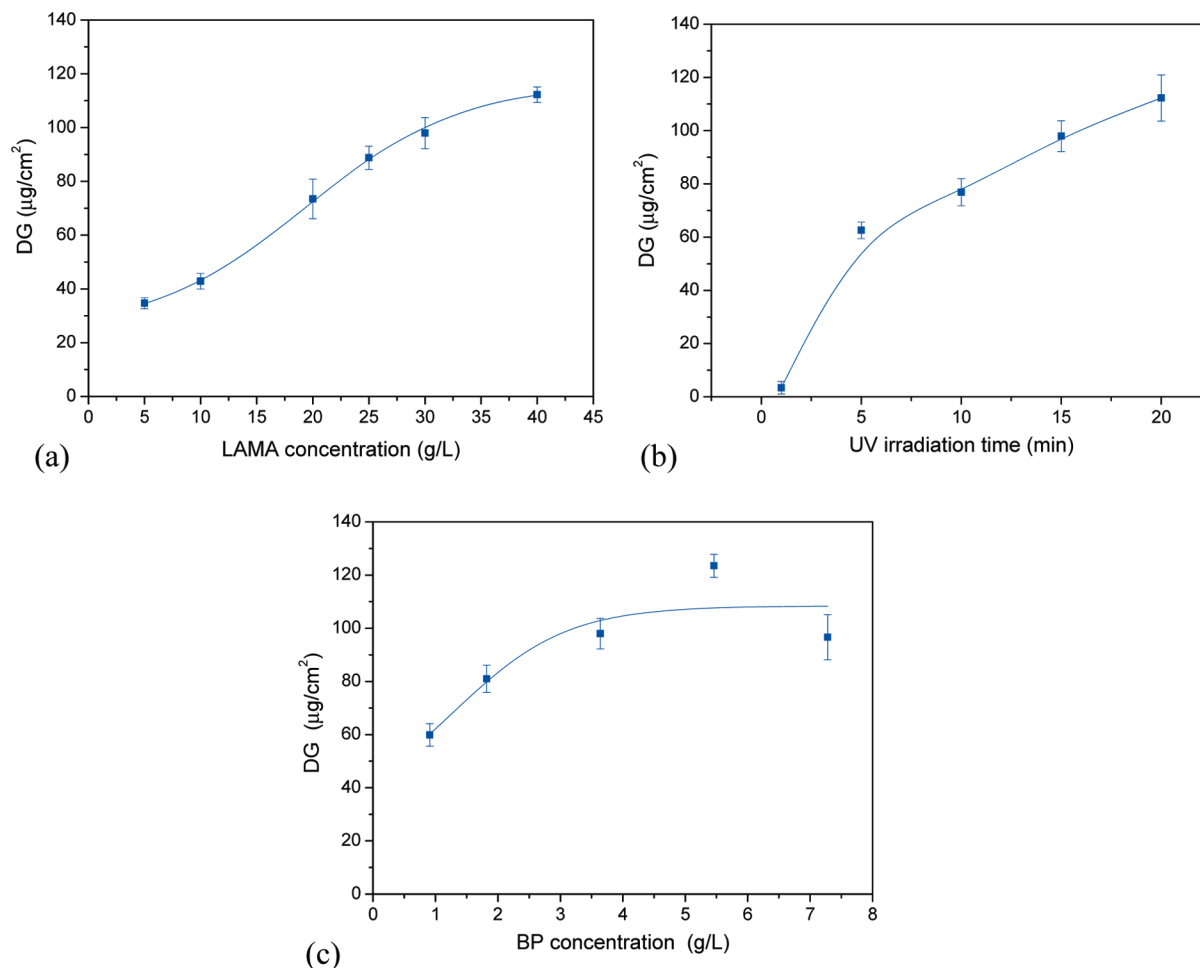


FIGURE 1. Effects of UV functionalization conditions on degree of grafting (DG): (a) monomer LAMA concentration (UV irradiation, 15 min; BP concentration, 3.64 g/L); (b) UV irradiation time (LAMA concentration, 30 g/L; BP concentration, 3.64 g/L); (c) photoinitiator BP concentration (LAMA concentration, 30 g/L; UV irradiation, 15 min).

with software. All results were an average of at least five measurements.

Evaluation of Bacteria Adhesion Specificity. Bacteria Adhesion. For this experiment, a bacterial cell density of 3×10^7 cells/mL was used for both *E. faecalis* and *S. maltophilia*. Bacteria were suspended in 10 mM HEPES buffer (pH 7.4) containing 0.1 mM Ca^{2+} which is essential for the interaction between galactose moieties and the sugar binding protein on bacterial cell membrane. Poly(LAMA) grafted membranes (approximately 4 cm^2) were immersed in a 45 mL bacteria suspension and incubated for 20 h at 30 °C. Then, the membranes were taken out and rinsed with HEPES buffer solution. After staining of bacterial cells using the DNA-binding fluorochrome 4',6-diamidino-2-phenylindole (DAPI), the membranes were observed under a fluorescence microscope. Unmodified PP membranes were treated in the same way as a control.

In some experiments, *E. faecalis* and *S. maltophilia* were used in mixed suspensions which contained 10% *E. faecalis* and 90% *S. maltophilia*. Experimental conditions and final total cell density detection were as described above. Differentiation between both strains during fluorescence microscopic evaluation of cell adhesion was done visually based on the significantly different cell shapes.

Competitive Sugar Inhibition of Bacteria Adhesion. In this experiment, a bacteria density of 3×10^7 cells/mL was used. After incubation in a bacteria suspension for 20 h at 30 °C in 10 mM HEPES buffer (pH 7.4) containing 0.1 mM Ca^{2+} as described above, the grafted membranes were incubated in 0.2 M sugar solution (D-(+)-galactose and D-(+)-glucose in HEPES

buffer) or plain HEPES buffer for 30 min at room temperature. Samples were then rinsed with HEPES buffer and stained with DAPI before fluorescence microscopic observation.

Influence of Primary Blocking of Glycopolymer by Lectin on Bacteria Adhesion. In these experiments, poly-(LAMA) grafted membranes were preincubated in lectin solutions (1 g/L in 10 mM HEPES buffer pH 7.4 containing 0.1 mM Ca^{2+}) for 30 min at room temperature. PNA was used as a specific binding lectin, and Con A and BSA were used as nonspecific controls. Afterward, the membranes were rinsed with HEPES buffer and were subsequently incubated in bacteria suspensions for 20 h at 30 °C as described above. Then, the membranes were taken out and rinsed with HEPES buffer solution. After staining of bacterial cells using the DNA-binding fluorochrome DAPI, the membranes were observed under a fluorescence microscope.

RESULTS AND DISCUSSION

Surface Grafting. Figure 1 shows the effects of monomer (LAMA) concentration, UV irradiation time, and benzophenone (BP) concentration on the degree of grafting (DG). As can be seen from Figure 1a, the DG increased with the increase of LAMA concentration. It seems that the trend of DG increase slowed down when the LAMA concentration exceeded 30 g/L. We tried to further increase LAMA concentration to prove this observation but the highest concentration we could achieve was 40 g/L. However, similar

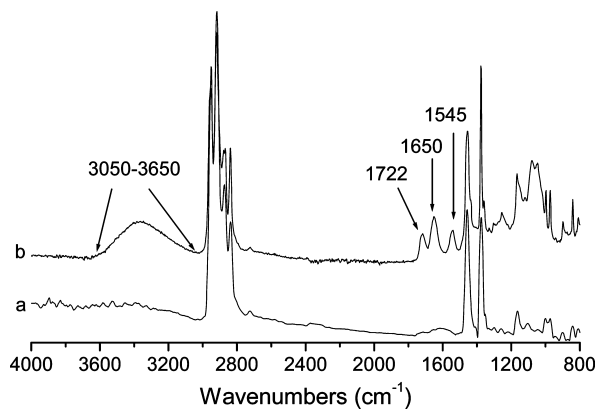


FIGURE 2. IR spectra of unmodified (a) and poly(LAMA) grafted (b) PP membranes.

phenomenon was also found when grafting another glycomonomer with monosaccharide side group to PP membrane surface (29). This may be ascribed to the increased viscosity at high monomer concentration which hindered the diffusion of monomer from the bulk solution to the growing chains at the membrane surface and to the extensive homopolymerization observed at higher monomer concentration. The effect of UV irradiation time on DG is shown in Figure 1b. The DG increased with the UV irradiation time in the range of 0–20 min. With the increase of UV irradiation time, more active radical sites were generated on the membrane surface and higher DG was obtained. Figure 1c shows the relationship between DG and BP concentration. A series of initiator solutions (BP in *n*-heptane) with different concentration were used, and the monomer concentration

and UV irradiation time were fixed at 30 g/L and 15 min, respectively, for this study. As can be seen, increase of initiator concentration can bring significant enhancement of DG because more active sites were generated and, subsequently, more polymer chains were tethered. In our experiment, heptane swelled the outer surface layer of PP membrane and, after drying/deswelling, this led to entrapment of BP in this layer. In the same incubation time, increase of BP concentration may lead to a saturation of amount of the entrapped initiator. Thus, further increasing the BP concentration failed to enhance DG, and DG reached a plateau.

Membrane Characterization. FT-IR/ATR was used to confirm the grafting of the poly(LAMA). Figure 2 shows the spectra of unmodified and poly(LAMA) grafted PP membranes. Compared with unmodified membrane, the poly(LAMA) modified membrane showed an absorption at 1722 cm^{-1} which can be attributed to the carbonyl groups in ester bond. Absorptions at 1650 and 1545 cm^{-1} belong to the amide I and amide II, respectively. Additional broad absorptions between 3050 and 3650 cm^{-1} due to NH and OH stretching vibration have also been found. These diagnostic absorption peaks in the IR spectrum indicate the grafting of poly(LAMA).

Morphological change of the poly(LAMA) grafted surface was detected by SEM. Representative SEM images are shown in Figure 3. As can be seen, the unmodified PP membrane showed relatively high porosity and small pore size (Figure 3a). However, after graft copolymerization of

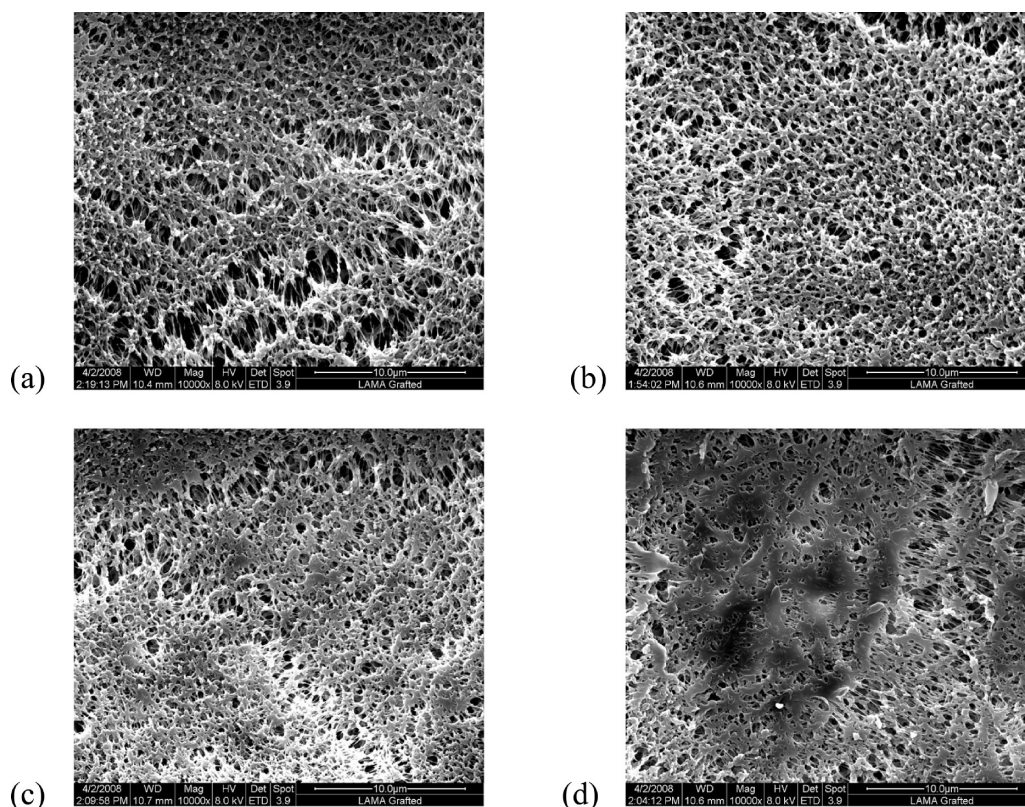


FIGURE 3. SEM images of the unmodified and poly(LAMA) grafted PP membranes: (a) unmodified, (b) DG = 40.8 $\mu\text{g}/\text{cm}^2$; (c) DG = 67.4 $\mu\text{g}/\text{cm}^2$; (d) DG = 136.7 $\mu\text{g}/\text{cm}^2$.

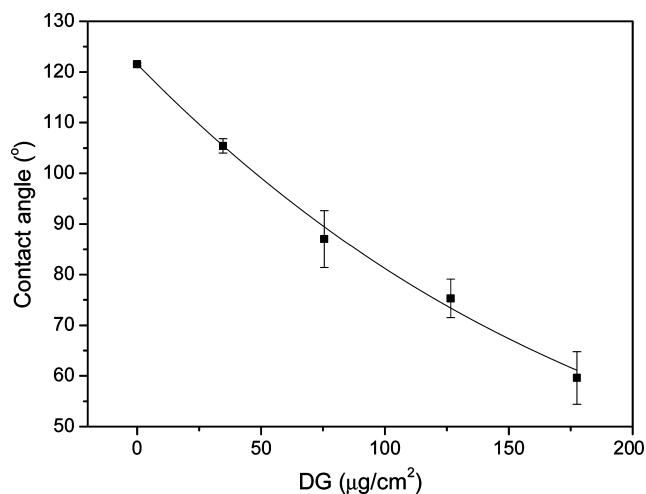


FIGURE 4. Water contact angles of unmodified and poly(LAMA) grafted PP membranes as function of the degree of grafting.

poly(LAMA), the surface was gradually covered by polymer and some of the pores were even blocked (Figure 3b–d).

Figure 4 shows the results of water contact angle measurements. The unmodified membrane was found to be highly hydrophobic and showed a contact angle higher than 120 degrees. With the grafting of poly(LAMA), the hydrophilicity was greatly enhanced and a significant decrease of contact angle down to approximately 60 degrees at the highest DG was observed. This could be ascribed to the hydrophilic nature of grafted poly(LAMA) chains which are abundant in hydroxyl groups.

Bacteria Adhesion. To investigate the capability of surface-tethered poly(LAMA) to select bacteria with galactose-

Table 1. Adhesion of *E. faecalis* and *S. maltophilia* to the Unmodified and Poly(LAMA) Grafted PP Membrane Surface

bacteria	cell density (cells/cm ²) ^a		increase factor ^b
	unmodified	poly(LAMA) grafted	
<i>E. faecalis</i>	$(8.8 \pm 2.0) \times 10^4$	$(3.4 \pm 2.3) \times 10^6$	39.1
<i>S. maltophilia</i>	$(6.2 \pm 1.0) \times 10^6$	$(7.4 \pm 1.3) \times 10^6$	1.2

^a Average values from three parallel samples, either unmodified or with DG of $165.7 \pm 4.2 \mu\text{g}/\text{cm}^2$. ^b Values obtained by dividing cell density on poly(LAMA) grafted membrane with that on unmodified membrane.

binding lectins or lectin-like proteins on their surface, we used *E. faecalis* and *S. maltophilia* to study their adhesion to unmodified and poly(LAMA) grafted PP membranes. *S. maltophilia* was used as a negative control to show the specificity of the interaction between poly(LAMA) and the lectins on the surface of *E. faecalis*. These strains were selected because it is known that *E. faecalis* has a galactose binding lectin on cell surface and *S. maltophilia* does not (14). The results of the bacteria adhesion assay (Figure 5 and Table 1) showed different adhesion behavior of *E. faecalis* and *S. maltophilia* onto unmodified as well as on poly(LAMA) grafted PP membranes. On the unmodified membrane, both strains adhered as single cells and the density of *S. maltophilia* was much higher than that of *E. faecalis* (Figure 5a,b, and Table 1). This difference in density of adhered bacteria on an unmodified membrane may be ascribed to the different nature of the two strains, e.g., cell surface properties.

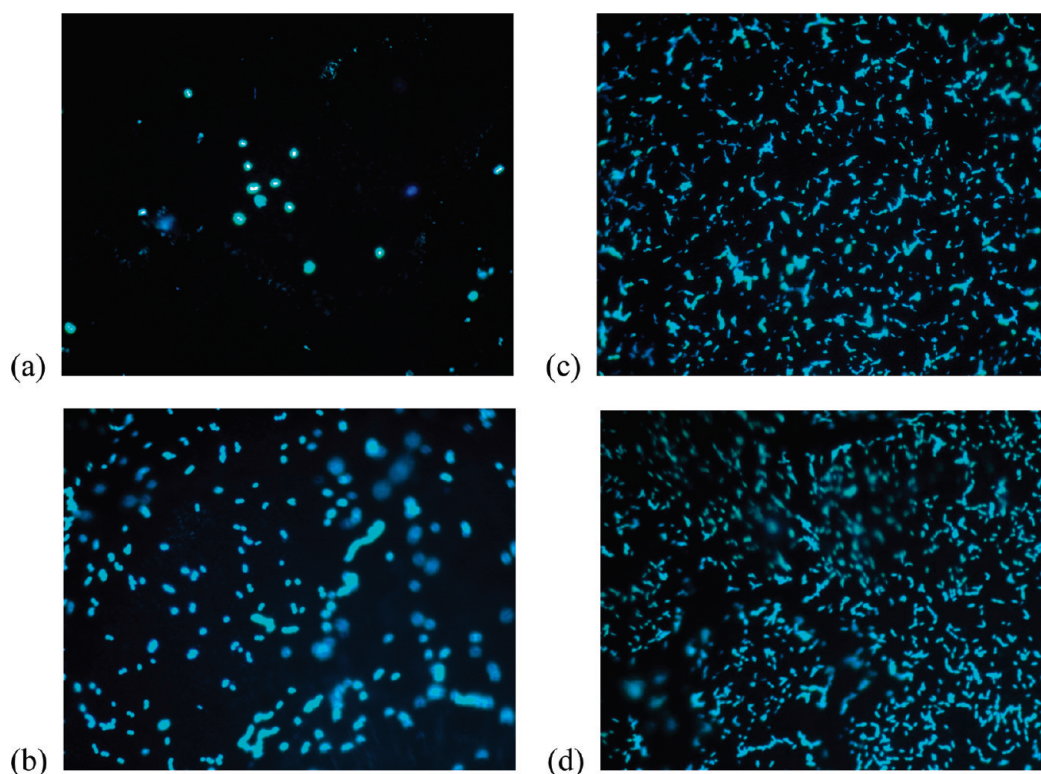


FIGURE 5. Fluorescence microscope images of DAPI stained *E. faecalis* and *S. maltophilia* on PP membrane surface. (a, b) Unmodified PP membrane with *E. faecalis* and *S. maltophilia*, respectively. (c, d) Poly(LAMA) grafted PP membrane ($\text{DG} = 63.3 \mu\text{g}/\text{cm}^2$) with *E. faecalis* and *S. maltophilia*, respectively.

Table 2. Adhesion of *E. faecalis* and *S. maltophilia* Mixed Cell Suspension to the Unmodified and Poly(LAMA) Grafted PP Membrane Surface

bacteria	cell density (cells/cm ²) ^b		increase factor ^c
	unmodified	poly(LAMA) grafted	
<i>E. faecalis</i> ^a	$(4.3 \pm 1.7) \times 10^3$	$(4.2 \pm 2.3) \times 10^4$	9.8
<i>S. maltophilia</i> ^a	$(5.8 \pm 0.7) \times 10^6$	$(4.8 \pm 0.6) \times 10^6$	0.8

^a 10% *E. faecalis* and 90% *S. maltophilia* mixed cell suspension.

^b Average value from three parallel samples, either unmodified or with DG of $165.7 \pm 4.2 \mu\text{g}/\text{cm}^2$. ^c Values obtained by dividing cell density on poly(LAMA) grafted membrane with that on unmodified membrane.

The adhesion to unmodified membranes is based on non-specific attractive forces between cells and solid surfaces (cf. ref 3).

In contrast, on the poly(LAMA) grafted surface, *E. faecalis* adhered with a significantly increased cell density (Figure 5c) which was 39 times higher than that on unmodified membranes (cf. Figure 5a). This indicates larger attractive forces to poly(LAMA) grafted than to unmodified PP. *S. maltophilia* adhered on the poly(LAMA) grafted surface in the same density as on the unmodified support (cf. Figure 5b,d), indicating no specific interaction between cell surface and support surface. This result for the negative control under identical experimental conditions confirms a specific recognition of *E. faecalis* by the poly(LAMA) grafted membrane.

To evaluate the specificity of this recognition between galactose residues on the poly(LAMA) grafted membrane surface and *E. faecalis* under more complicated conditions, a competition adhesion experiment was conducted. The same procedure as for primary adhesion was taken with the sole modification of the use of a mixed bacteria suspension which contained 10% *E. faecalis* and 90% *S. maltophilia*. Results are shown in Table 2. With only 10% content in the mixed cell suspension, *E. faecalis* had a much lower density than that in a primary adhesion experiment. Nevertheless, similar to the primary adhesion experiment with the single strain, *E. faecalis* exhibited an increased, about 10 times, cell density on the poly(LAMA) grafted surface whereas the density of *S. maltophilia* was almost the same on poly(LAMA) grafted and unmodified PP membranes. However, this increase was lower than that in primary adhesion which may be explained by the lower cell density of *E. faecalis* in the

mixed bacteria suspension. These results demonstrated that, even in the presence of another bacterial strain, the galactose residues on membrane surface were still specifically attractive to *E. faecalis*. Moreover, these results also indicate the possibility of applying poly(LAMA) grafted PP membrane for capture under more realistic operation conditions, e.g., from natural water which contains various bacteria strains.

Competitive Sugar Inhibition of Bacteria Adhesion. The interaction between sugar binding proteins such as lectins and glycoconjugates can be inhibited by the free sugar for which the lectins are specific (10). For applications, e.g., in a diagnostic system, it may be also important to regenerate the immobilized receptors. Galactose is the sugar specific for the lectins exhibited on the *E. faecalis*; thus, we investigated if the adhesion of *E. faecalis* to the poly(LAMA) grafted membrane could be inhibited by concurrent binding to galactose added to the samples. Therefore, after 20 h of primary adhesion of *E. faecalis*, the modified membranes were washed to remove bacteria that were not adhered and subsequently incubated with different sugar solutions for 30 min. Figure 6 shows the fluorescence microscope images of adhering *E. faecalis* after addition of 200 mM galactose, 200 mM glucose, and only buffer without sugar. In the positive control experiment, after incubation in buffer without sugar, 8.9×10^6 cells/cm² adhered on poly(LAMA) grafted membranes. After incubation with the specific sugar galactose, only 1.4×10^6 cells/cm² were counted on the poly(LAMA) membranes. Thus, a significant decrease in cell adhesion could be observed due to the presence of the inhibition sugar. Interestingly, after incubation with the nonspecific sugar glucose 1.5×10^7 cells/cm² were adhered. The slightly higher cell density on glucose treated sample compared to the sample without sugar could be due to the experimental error. However, this result indicates that glucose has no inhibition ability to the interaction between galactose residue on poly(LAMA) chains and *E. faecalis* cells. Anyway, the 6- to 10-fold decreased cell density on grafted poly(LAMA) after short incubation with galactose can be explained with the competition between free galactose and galactose residues on poly(LAMA) for bacterial binding sites causing a detachment of previously adhered *E. faecalis*. Further, it confirms the highly selective recognition of *E. faecalis* by surface tethered poly(LAMA).

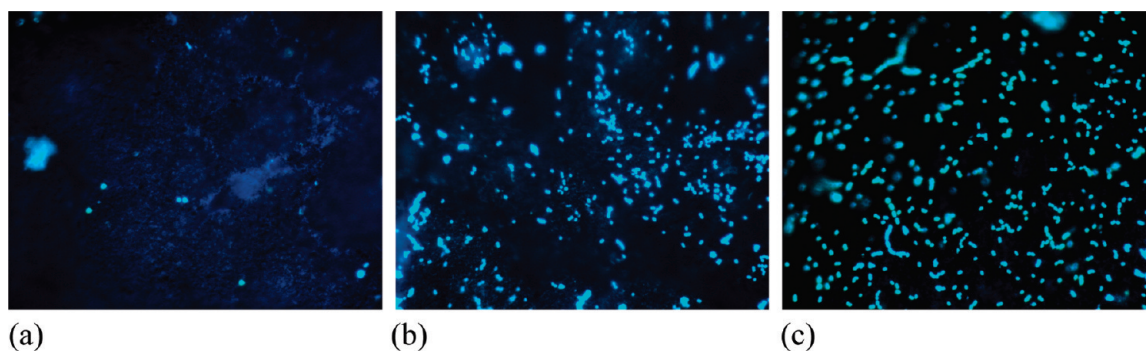


FIGURE 6. Fluorescence microscope images of adhering *E. faecalis* (DAPI stained) on poly(LAMA) grafted PP membrane (DG = $106.1 \mu\text{g}/\text{cm}^2$) after 30 min inhibition with (a) 200 mM galactose in buffer, (b) 200 mM glucose in buffer, and (c) sugar-free HEPES buffer solution.

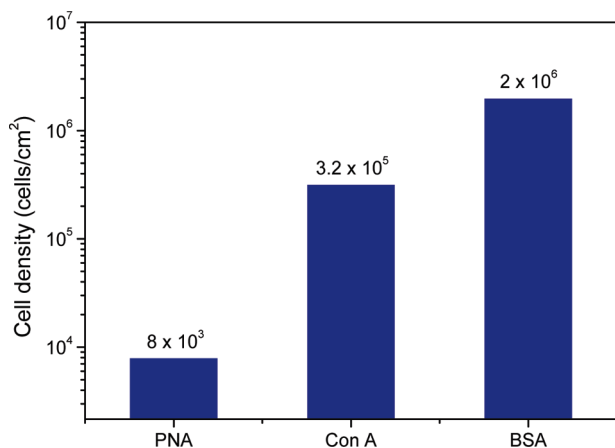


FIGURE 7. Adhesion of *E. faecalis* after primary adsorption of PNA, Con A, and BSA to a poly(LAMA) grafted PP membrane (DG = 110.6 $\mu\text{g}/\text{cm}^2$).

Influence of Primary Blocking of Glycopolymer by Lectin on Bacteria Adhesion. To further confirm the specificity of the bacterial binding in the case of *E. faecalis*, a lectin blocking experiment was carried out. PNA is a lectin which can specifically recognize galactose (10, 36). We employed PNA to investigate if the sugar residues on the membrane surface can be blocked by this lectin and prevent following adhesion of *E. faecalis*. The nonspecific protein BSA and Con A, another lectin which is not specific to galactose, were used as negative controls in this investigation.

As shown in Figure 7, PNA has significant blocking effect. After adsorption of PNA to the poly(LAMA) grafted surface, most galactose residues on the membrane surface were occupied and only 8.0×10^3 cells/cm² were achieved for *E. faecalis* adhesion. However, grafted membranes pretreated by adsorption of BSA showed much higher *E. faecalis* adhesion (2.0×10^6 cells/cm²); the value was similar to data without attempted protein blocking (cf. Table 1). This indicated that much less BSA was adsorbed on the surface. BSA is known as a protein adsorbing nonspecifically to hydrophobic surfaces and having no special interaction with any sugar. Thus, in our case, poly(LAMA) grafted PP membrane surface was not attractive to BSA due to the hydrophilic nature of glycopolymer and to the fact that no significant attractive interaction between BSA and galactose was possible. Therefore, sugar residues were still available for bacteria after incubation with BSA. On the other hand, no significant blocking effect was also found on Con A primarily adsorbed membranes, but cell density was lower than in case of BSA. Con A can be specifically recognized by glucose which differs from galactose only in the configuration of one hydroxyl group. From the glycobiology we know, the configuration of hydroxyl groups on the saccharide ring is vital for the carbohydrate–protein interaction (10, 37, 38). Although the configuration changes of one or two hydroxyl groups lead to the lack of some hydrogen bonds, the interaction could still happen in a weaker way. For this reason, Con A may form weak hydrogen binding with galactose and adsorb to the membrane surface. Despite that most of the Con A had been washed off the surface in the

rinsing step, a considerable amount of galactose residues were still occupied which resulted in the slight suppression of bacteria adhesion.

CONCLUSIONS

In conclusion, in this study, we demonstrated a grafted synthetic glycopolymer on the surface of a support micro-filtration membrane showing a pronounced and selective interaction with the *E. faecalis* cell surface leading to specific bacteria adhesion. A glycomonomer bearing galactose as side group (LAMA) had been grafted to a PP membrane surface by UV-induced graft copolymerization. With grafted poly(LAMA) on the surface, adhesion of *E. faecalis* was enhanced by about 40 times in comparison with unmodified membrane while *S. maltophilia* showed almost the same cell density on both surfaces. Similar results were also obtained in a competition experiment using a mixed bacteria suspension of both strains. Further, the adhered *E. faecalis* cells could be released by incubation with galactose solution which gives the possibility to regenerate the receptors on the membrane surface and, on the contrary, glucose solution showed no inhibition effect. Occupying the sugar residues primarily by PNA, a lectin specifically binding to galactose, the adhesion of *E. faecalis* was significantly suppressed. At the same time, BSA and Con A exhibited no or only a very weak inhibition effect on *E. faecalis* adhesion, respectively. These results demonstrate that *E. faecalis* can be selectively captured by poly(LAMA) grafted PP membrane and that this capture is based on the interaction between galactose residues on the membrane surface and the galactose-binding receptors on the bacterial cell membrane. Our results also indicate that this surface-tethered multifunctional receptor-like glycopolymer is very promising for selective binding of bacteria from more complex mixtures. Depending on the pore size of the support, it would be possible to capture bacteria from aqueous streams in tangential flow or flow-through systems for analytical, water treatment, or biomedical applications. It can be expected that, by varying the pendant sugar groups, with the respective specific sugar receptor mixture in the grafted polymer layer, it would be also possible to select different kinds of bacteria at the same time. Further ongoing research focuses on evaluating long-term effects of grafted poly(LAMA) on adhering *E. faecalis* and on performing bacteria capture from flowing aqueous streams.

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