Introduction Elucidation of the mechanisms of bacterial adhesion is critical for a better understanding of the bacterial-host interactions. Bacterial adhesion can be mediated via either receptor-ligand interactions or non-specific hydrophobic and electrostatic interactions. Bacterial initial adhesion to a surface is important as it is the first step in biofilm formation on medical implants and is believed to be responsible for biomaterial-centered infections (1). Besides the non-specific interactions, gram-negative bacteria can also use the lectins on their fimbriae tips to specifically interact with the carbohydrates on cell surfaces (2). In this research, our aim was to compare the different interactions during bacterial adhesion to a surface through use of the rapid and highly sensitive QCM-D technique (Figure 1).
Materials and Methods

We have synthesized glycopolymers using Reversible Addition-Fragmentation Chain Transfer (RAFT) polymerization which were subsequently immobilized on QCM-D sensor surface for the studies of bacterial adhesion. Ricinus communis Agglutinin (RCA120), a galactose specific lectin, was first studied by QCM-D to determine the specific lectin interactions to the different glycopolymer-treated surfaces. Subsequently, Pseudomonas aeruginosa PAO1 (a gram-negative bacterium with galactose specific binding lectin (PA-IL) (3)) and Escherichia coli K-12 (a gram-negative bacterium with mannose specific binding lectin (4)) were used as model bacteria to study bacterial adhesion mechanisms on different polymer-treated sensor surfaces by the coupled resonance theory.

Results

The polymers were synthesized by the RAFT technique. The molecular weights (around 13 kDa) and molecular weight distributions (PDI) (smaller than 1.4) were determined by gel permeation
chromatography (GPC). Polymers with dithioester terminal groups were able to attach on the gold coated QCM-D sensor chip surfaces as indicated by the negative QCM-D frequency shifts. The interactions of lectins on glycopolymer-modified surfaces were selective due to the specific carbohydrate-lectin interactions. Although a higher number of bacteria were found to adhere on the cationic polymer surfaces via the electrostatic interaction, significant number were found dead due to the antibacterial properties of the amine based cationic polymer. A significant higher amount of P. aeruginosa PAO1 was found to adhere on the galactose containing glycopolymer surface with strong bond stiffness by QCM-D (Figure 2), as compared to E. coli K-12 on the same surface (Figure 3). The adhesion of P. aeruginosa PAO1 to the glycopolymers were also highly dependent on the presence of calcium ions (Figure 2).

**Discussion and Conclusion**

In this study, we provided the first example of using glycopolymers to study specific bacteria-substratum interactions by QCM-D technique. Three factors
were considered during the investigation of bacterial adhesion such as the bacterial lectins, the carbohydrate residues, and the test media. Interesting positive frequency shifts for C-type lectin (PA-IL) containing bacteria (P. aeruginosa PAO1) adhering on galactose containing polymer (PLAEMA) surface in 10 mM CaCl2 were interpreted by coupled resonance model. By comparing to the results on bacterial adhesion on cationic polymer surface by non-specific electrostatic interaction, we believed these positive frequency shifts might relate to the stronger bond stiffness between bacterial lectin and carbohydrate residues on the glycopolymers due to the “glycoside cluster effect”.

Figure 1. Schematic representation of the specific interactions of bacteria to glycopolymers immobilized on QCM-D surface based on the coupled
resonance theory.

Figure 2. Frequency and dissipation shifts of P. aeruginosa PAO1 adhesion on different polymers treated QCM-D sensor surface (in 10 mM CaCl2). a: PGAPMA, b: PLAEMA, c: PEG-SH. d and e: Calcium dependence of P. aeruginosa PAO1 adhesion on PLAEMA surface.
Figure 3. Frequency and dissipation shifts of E. coli K-12 adhesion on different polymers treated QCM-D sensor surface (in 10 mM CaCl2). a: PGAPMA, b: Glu, c: Gal, d: PEG.
PLAEMA, c: PEG-SH.

References