Introduction
This application note describes QCM-D data obtained during the formation of a polysaccharide polyelectrolyte multilayer. It is an example of a biomedical application of the polyelectrolyte multilayer approach. These applications are attractive due to their ease of preparation and the fine control over the material structure. A high degree of versatility is also offered, including the capability of incorporating high loadings of different types of biomolecules in the films and adjusting the film robustness under ambient and physiological conditions. In particular, natural polysaccharides, such as alginate and chitosan, have attracted interest since they resemble components in the extracellular matrix and show good biological properties while being accessible to chemical modifications.

Here, polysaccharide multilayers were created following a protocol described by Alves et al. Based on alternating deposition of the positively charged polysaccharide chitosan and the negatively charged polysaccharide alginate onto gold coated QCM-D sensors. The resulting multilayer structure is schematically shown in figure 1 together with the chemical structures for the polysaccharides. The experiments were performed using an automated QCM-D setup (Q-Sense E4 Auto) allowing 18 h long measurements including a terminal cross-linking step of the chitosan/alginate multilayer.

Results and discussion
The QCM-D frequency and dissipation shifts during the build-up of 31 alternating layers of the polysaccharides chitosan and alginate are shown in figure 2. Each layer deposition can be distinguished by stepwise decrease and increase in frequency and dissipation, respectively. Mass changes are related to changes in oscillating resonance frequency, $\Delta f$, where a negative frequency shift is equivalent to mass increase. The dissipation, $\Delta D$, is related to the energy dissipation of the crystal oscillation, and a positive shift indicates a less rigid structure. The relatively small ratio between the dissipation and the frequency shifts suggests a continuous build-up of a quite rigid and compact polyelectrolyte multilayer as is expected for these highly charged polysaccharide chains. This rigidity is also reflected in the small separation of the
frequency overtones. The measurements were performed using the E4 Auto setup. The system proved great stability over the entire measurement sequence of 18 hours under constant flow conditions and the magnitudes of each of the deposited layers increased during the first 6-7 layers to become close to linear for the additional layers. The characteristics of the deposition were similar to the published data on this system including the terminal cross-linking step with glutaraldehyde 1. The action of the cross-linking agent gave only a small dissipation shift and close to no frequency shift. These small responses were rationalized in the study by Alves et al, where infrared spectroscopy showed that only the top chitosan layer was cross-linked. Cross-linking is an attractive way of controlling the stability of polyelectrolyte multilayers, enabling for example control of cell adhesion, wettability or release of incorporated biomolecules.

Conclusions
Polysaccharide multilayers of chitosan and alginate were successfully built up using the Q-Sense E4 Auto setup for 18 h measurements while allowing minimization of the hands-on time to approximately 1 hour. The obtained results proved close to linear build up of 31 layers of a quite rigid structure. Cross-linking at the end of the experiment had little effect on the QCM-D responses and probably only occurred in the top chitosan layer. This study shows the E4 Auto as a reliable and efficient tool for long-time measurements and enables experimental setups that would have been cumbersome to perform by manual sample handling.

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References: