CHAPTER I

INTRODUCTION

1.1 Statement of Problem

The performance of biomedical devices and biomaterials depends greatly upon the surface properties of the materials since it is the surface of the materials that first comes into contact with their biological surroundings. Several approaches have been developed for the modification of material's surface to suit specific biomedical applications. The layer-by-layer (LBL) assembly technique [1], which is based on the alternating adsorption of oppositely, charged polyelectrolytes, is an alternative strategy for film formation that can be used for biomedical applications. This versatile technique mostly involves building up of thin multilayered polyelectrolyte film. The electrostatic interactions between a charged polyelectrolyte and an oppositely charged surface lead to the polyelectrolyte adsorption from aqueous solution and to the surface charge reversal, enabling a second adsorption of an oppositely charged polyelectrolyte. The build-up is easy and procedure can be adapted for any type of surface as long as the surface charge is present and without the shape limitation. The thickness of multilayer film can be varied as a function of adsorption variables as well as the number of deposition. Most importantly, the surface properties of the supported films are determined by the outermost surface layer that is last deposited.

The use of natural biomaterials is an attractive concept for the development of polyelectrolyte multilayers. One of the biomaterials for this application that has attracted most interest is chitosan. It is the partially deacetylated form of chitin, the second-most abundant natural polymer, mainly harvested from the exoskeleton of insects, marine crustaceans and fungal cell walls. It is favorable physico-chemical and biological properties such as biocompatible [2], biodegradable [3,4], non-toxic [5], and antibacterial activity [6] that makes chitosan a suitable biomaterial.

Up to now, although several researches have been reported on the formation of polyion complex multilayer film between chitosan and a number of anionic polyelectrolytes, none has been mentioned on charged derivatives of chitosan whose biological properties are significantly different from those of chitosan. Quaternary ammonium chitosan, a positively charged derivative possesses superior antimicrobial activity [7,8] whereas succinyl chitosan, a negatively charged derivative of chitosan, has favourable properties such as low toxicity and long-term retention in the body so it was applied as a drug carrier [9], and sulfonated chitosan, a negatively charged derivative of chitosan, is antithrombogenic [10,11].

Our previous study [12], has demonstrated that the multilayer films assembly of charged derivatives of chitosan (CHI), *N*-sulfofurfuryl chitosan (SFC) and *N*-[(2-hydroxyl-3-trimethylammonium)propyl]chitosan chloride (HTACC) can be successfully built up on the plasma-treated PET. Alternate bioactivity of the multilayer assembly of three pairs of polycation-polyanion CHI- poly(sodium styrene sulfonate) (CHI-PSS), poly(allylamine hydrochloride)-SFC (PAH-SFC), and HTACC-poly(acrylic acid) (HTACC-PAA) was tested against four proteins (albumin, fibrinogen, γ-globulin, and lysozyme). It has been demonstrated that these two charged derivatives of chitosan can be potential candidates in biomedical-related applications. From a practical point of view, it is also important to evaluate the cellular responses of these different polyelectrolyte multilayer films.

In the present study, we focus our attention on assembling polyion complex multilayer films from three pairs of oppositely charged polyelectrolytes: *N*-[(2-hydroxyl-3-trimethylammonium)propyl]chitosan chloride (HTACC) and poly(acrylic acid) (PAA) (HTACC-PAA), poly(allylamine hydrochloride) (PAH) and *N*-succinyl chitosan (SCC) (PAH-SCC), and poly(allylamine hydrochloride) (PAH) and *N*-sulfofurfuryl chitosan (SFC) (PAH-SFC). The LBL film growth was monitored by quartz crystal microbalance (QCM), atomic force microscopy (AFM), water contact angle measurement, attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR). The biological responses of all polyion complex multilayer films were investigated by cell adhesion and cell proliferation of fibroblasts using an established mitochondrial activity (MTT) assay. We hypothesize

that an alternate response can be achieved as long as each layer is thick enough and overall biological response depends on the outermost layer. The consequence of this study should provide fundamental information that can be useful for further development of these charged chitosan derivatives for biomedical applications.

1.2 Objectives

1. To prepare polyion complex multilayer films containing charged derivatives of chitosan using layer-by-layer assembly.
2. To study cellular responses of polyion complex multilayer films containing charged derivatives of chitosan.

1.3 Scope of Investigation

The stepwise investigation was carried out as follows:

1. Literature survey for related research work.
2. Synthesis of positively and negatively charged derivatives of chitosan.
3. Investigation of the effects of adsorption variables on the individual and overall thicknesses of assembled film.
4. Preparation of multilayer films from charged derivatives of chitosan.
5. Characterization of multilayer films.
6. Determination of biological response of multilayer films with fibroblasts.