SUMMARY

The biomolecule patterning methods have been very important in bioanalysis lately, because they are applied in the fabrication of biomolecule microarrays, which are components of the contemporary bioanalytical devices. Although photoresist photolithography is a successful thin film patterning method, it couldn't be used in biomolecule patterning until now, because of the non-biocompatibility of the lithographic conditions and materials. Thus, biomolecule patterning has been achieved mainly with the *nitrobenzyl photochemical method* and with two *delivery methods*.

The *object of this dissertation* was the research and development of appropriate photolithographic process and photoresists for protein patterning on substrate. The approach of this goal was done in five stages.

Initially, *new photolithographic process was proposed* for application in protein patterning. It is a *modified version of the lift-off photolithography* used in metallization in microelectronics. This version differs from its classic one in the biocompatibility of the lithographic conditions, the photolithographic removal of the remaining photoresist film and the addition of one step (blocking) after each protein deposition in relation to the metal deposition.

Afterwards, *the requirements the photoresists should fulfill* in order to be appropriate for the chosen photolithographic process were determined. These requirements are divided in *biocompatibility requirements, repetition requirements and requirements posed by the substrate material and geometry.* The biocompatibility requirements which are the most important ones, are the following three: (1) exposure in visible or near UV, (2) thermal treatment in temperature less than 50^{o}C, and (3) development in standard aqueous base solution AZ (aqueous TMAH solution 0.26 N) diluted at least 100 times with deionized water.

In a following stage, *candidate photolithographic materials* characterized by *two different dissolution change mechanisms*: (1) *photooxidation mechanism*, and (b) *photoacid generation and acidic catalysis mechanism (chemical amplification mechanism),* were studied. In the photooxidation mechanism materials the photooxidation of poly(2-hydroxylethyl methacrylate) (PHEMA) by polyoxometallic anions and molybdenum(V) porphyrins was examined. In the first case the 12-tungstenphosphoric acid was indeed capable to photooxidize PHEMA in biocompatible conditions, but it was dissolved by the water. In the second case, although molybdenum(V) porphyrins had remarkable photooxidation behavior in chlorinated solvents, they couldn't photooxidize PHEMA in photopolymeric films.

In the chemical amplification mechanism materials, firstly acrylic homopolymers and then (meth)acrylic copolymers, having as photoacid generator one of the sulfonium salts (the triphenyl sulfonium salt, TPS, and a specific triaryl sulfonium salt, TAS) were studied. The dissolution change reaction mechanism in these materials, which was similar to the "tBOC" chemical amplification photoresist in 248 nm, was the acid catalyzed cleavage of the tert-butyl pendant polymer group during the postexposure thermal treatment. From the acrylic homopolymers, poly(*t*-butyl acrylate) (PTBA) and poly(*t*-

butyl methacrylate) (PTBMA) were examined, while from the (meth)acrylic copolymers poly(2-hydroxylethyl methacrylate-*co*-isobornyl methacrylate-*co*-*t*-butyl methacrylate-*co*-acrylic acid) (MAC1) with initial weight ratio of monomers 30:40:20:10, and poly(2-hydroxylethyl methacrylate-*co*-isobornyl methacrylate-*co*-*t*-butyl methacrylate-*co*-methacrylic acid) (MAC2) with initial weight ratio of monomers 30:34:25:11, were also studied. From the acrylic homopolymers, PTBA-based photoresist had the highest photosensitivity and could be processed in biocompatible lithographic conditions, but it suffered from a serious problem: the remaining of a thin photoresist layer caused by excessive exposure, which made difficult the performance of following lithographies. This is the reason why the two preceding (meth)acrylic photoresists were studied. From these materials MAC1-based photoresist had much higher photosensitivity (at least one order of magnitude) in relation to the PTBA photoresist and could be processed lithographically in even milder conditions: complete abolition of the postexposure thermal treatment and development in more diluted than 1:100 standard aqueous base solution AZ.

According to their photolithographic properties, *the most appropriate photoresists were chosen* for the photolithographic patterning of proteins on substrate. These photoresists, which were chemical amplification ones, were initially the photoresist based on the homopolymer PTBA and afterwards, as an evolution of it, the one based on the copolymer MAC1, with photoacid generator in both cases one of the two preceding sulfonium salts. The concentrations of polymer and photoacid generator in these photoresists were optimized, as well as the lithographic conditions.

In the end, *the results of protein patterning* using the proposed photolithographic process and the two chosen photoresists were *presented*. Although with PTBA photoresist only two proteins were patterned onto capillary inner surface, mouse IgG (MγG) and bovine serum albumin (BSA), the results with the MAC1 photoresist were much more important. Thus, two proteins, rabbit IgG (RγG) and BSA, were patterned using MAC1 photoresist in 2.5 μm least dimension microstructures on silicon substrate. Furthermore, three proteins, MγG, biotinylated bovine serum albumin (B-BSA) and BSA, were patterned using this photoresist in 6.25 μm least dimension microstructures on silicon substrate. Finally, two proteins, RγG and BSA, were patterned with this photoresist onto capillary inner surface.

It is believed that the above results, and generally the whole approach of this issue, contribute to the exhibition and establishment of photoresist photolithography as a reliable method for protein patterning, or at least they constitute the beginning to this direction.