Prague, April 12th, 2024

Review Report on the Ph.D. thesis of ***Yu-Min Wang,***

entitled

Macromolecular avenues for the creation of bio-inspired hierarchically structured surfaces

The research reported in the submitted Ph.D. thesis represents an original contribution to the emerging field of polymer biomaterials and surface science. The study focuses on the synthesis of dense polymer brush coatings on inorganic surfaces, with the aim of reducing their fouling by plasma proteins. The polymer brush coatings consisting of hydrophilic biocompatible polymers (mainly PHPMA) were synthesized by grafting-from as well as grafting-to approaches, with a comparison of physicochemical and antifouling properties obtained from both methods. Furthermore, the antifouling properties and hemocompatibility of polymer brush coatings by different polymer types were compared. Finally, 3D-printed scaffolds based on PHPMA have been developed.

The results included in this dissertation demonstrate the student's ability (i) to synthesize polymer brush-coated surfaces using RAFT by both grafting from and grafting to methods, as well as to characterize the prepared polymers and surfaces by numerous experimental techniques. (ii) to measure and understand the antifouling properties of polymer coatings, (iii) to evaluate potential applications of prepared materials.

The dissertation contains original research work and is based on one published article (Chapter 4.1.), with the candidate as the first author. Furthermore, one more manuscript is currently under revision, with expected acceptance soon.

The thesis is mostly well structured and correctly presented, with the main scientific data concentrated in chapters 4.1 and 4.2. Unfortunately, the orientation in the thesis manuscript is relatively difficult due to missing page numbers. Also, the subchapters of the R&D section beyond 4.3 are not well interconnected. This is true mainly for Chapter 4.5 (preparation of hydrogel scaffolds), with the research topic clearly different from the rest of the thesis. This chapter could have been easily omitted, but the thesis would still have significant scientific value. The thesis is written in good English with occasional typos. The thesis contains an appropriate number of citations, with occasional places with missing references (e.g., the comparisons with ATRP in chapter 4).

At the beginning of the thesis, an extensive literature review introduces the reader to the field of biomaterials and antifouling properties, as well as essential terms studied in the thesis, such as the structural and physical parameters of polymer brushes. Unfortunately, the thesis practically lacks any introduction to surface polymer chemistry. In such synthesis-oriented work, I would expect a significant part of the Introduction section to be devoted to a review of surface-initiated controlled radical polymerization techniques, a comparison of their benefits and drawbacks, and summarizing the work of other researchers on the topic. On the other hand, a substantial part of the introduction is devoted to extracellular matrix and 3D bioprinting, which is only merely mentioned in the experimental work. Finally, the Objectives section of the thesis is very general and does not contain any concretely defined aims.

In the Experimental section, the experiments are fully described, with several minor issues, such as missing yields in organic syntheses or incorrect structure of substituted methacrylamide in Scheme 3.

The following chapter 4 has been devoted to the own original research – with the implicit aim of optimizing the conditions to obtain polymer-coated surfaces with the best antifouling properties. To me, the main scientific highlights of the chapter that deserve appreciation are:

1. Optimization of SI-RAFT of HPMA, revealing correspondence of molar masses of SI and solution-born polymers
2. Finding that grafting-to in poor solvent leads to higher grafting density compared to coating in a good solvent
3. Characterization of surfaces by unique techniques, such as SMFS-AFM and neutron reflectometry.

The obtained results were arranged in eye-catching graphs and were well-discussed. The overall quality of the research is very high, and I have just several questions for the defense:

1. Within the manuscript, there is practically no discussion about the statistical significance of differences for the antifouling measurements, especially in comparing GT coating in GS and PS. Can you please comment on the statistical significance for Figures 15, 25, and 27?
2. In Figures 17 and 20, the kinetic graphs show only single points, while the SEC data show averages with error bars. Why?

3. Page 95: Statement: "The presence of CTA within a polymeric scaffold introduces the possibility of self-healing properties." Can you, please suggest a mechanism for such self-healing?
4. The author converts the chain-end CTA to free thiol for SMFS-AFM. Would it be possible to measure SMFS on brushes with retained CTA (they bind to gold, as well)?
5. Figure 25 compares the antifouling properties of different brushes at constant polymer DP 50, which, however, leads to different brush thicknesses. Could you assume the outcome of such a comparison for same-thickness coatings?
6. The prepared GT brushes contain chain-end carboxylic acid, which will be negatively charged at physiological conditions. On the other hand, GF brushes will remain neutral. Could this somehow influence the antifouling properties of brushes?
7. Could you, please show the mechanism of PET RAFT polymerization using TPO-L? To me, it looks more like photoinitiated polymerization and not PET-RAFT polymerization.
8. In the description of Figure 17, it is stated that the polymers have "narrow dispersity". However, from the graph, it is clear that some of the polymers have higher dispersity (>1.5). Could you, please explain to us the term "narrow dispersity" (i.e., the threshold)? Also, the dispersities of standard solution RAFT polymerizations are generally much lower (below 1.2) than in your solution-born polymers. Can you explain the difference?
9. In the thesis, PHEMA was used to immobilize PNA to the surface. Why have you used PHEMA when, essentially, the whole thesis focuses on PHPMA?

To summarize, the dissertation represents high-level scientific work in the area of surface polymer chemistry. All experiments have been well arranged; all measurements have been correctly applied. The scientific level of the thesis can be rated as excellent, while the presentation and format of the thesis can be rated as sufficient.

In my opinion, the reviewed thesis fulfills all the requirements posed on these aimed at obtaining a Ph.D. degree. Therefore, I can recommend this thesis to be defended in front of the respective committee.

RNDr. Ondrej Sedlacek, Ph.D.