Reviewer Report on PhD Thesis of Veronika Švachová

"Electrospinning of Modified Biopolymers for Medical Applications"

In her PhD thesis, Veronika Švachová investigated electrospinning of gelatin-based polymer nanofibers using Nanospider^{TM} NS LAB 500S machine from Elmarco s.r.o. In the 2nd chapter of the thesis the author provided a literature overview of electrospinning investigation and development. Several parameters (volatility of solvent, relative humidity, polymer solution properties, applied voltage, concentration of polymer solution, viscosity, surface tension and conductivity of polymer solution) were discussed to influence the electrospinning process according to the published results. The author commented on the scattered opinions that "According to literature, scientists disagreed on the same critical parameters." Despite, I am missing more synthesized discussion of the influential parameters supported also by some physical and/or chemical principles behind the effects.

The important part of the 2nd chapter is the state-of-the-art section describing hemostatic biopolymers, oxidized cellulose, collagen and gelatin, and their electrospinning. It reveals that oxidized cellulose (OC) have never been directly electrospun because of poor solubility and that the electrospinning of collagen can be just an expensive way how to produce gelatin fibers. These conclusions shows the challenges and directions of the experimental part of the thesis. The last part of the 2nd chapter is devoted to halloysite nanotubes (HNTs) and their polymer composites since the preparation of halloysite filled polymer nanofibers belonged to one of the aims of the thesis.

The experimental section of the thesis gives a short overview of electrospinning conditions and methods of sample characterization. The information on pages 46-47 does not seem to agree well with the Results and Discussion in which a wider range of conditions was used (15 and 12 cm on page 50; 6, 7, 9 and 12 wt.% of gelatin solutions on page 50). Mixing the units of concentrations of gelatin in acetic acid (wt.% versus v/v%) is confusing and does not allow clear understanding of the effect of parameters on the electrospinning of gelatin. The comparison of nanofiber diameters between different samples is several times performed without giving the average value and its error (or comparing the diameter distributions). Therefore, it is difficult to judge how significant are the differences between the samples.

Besides by SEM, the prepared materials were also investigated by infrared (IR) spectroscopy. The position of amide I and amide II bands was found to shift from 1634 to 1631 cm^{-1} and from 1528 to 1537 cm^{-1}, respectively, in gelatin (Gel)/sodium salt of oxycellulose (NaOC) in comparison with Ref. [300] (page 54) whereas the positions of 1645 and 1535 cm^{-1} were reported for the pure gelatin nanofibers on page 52. I think it needs some clarification, especially because the cited paper does not seem to discuss the position of amide peaks in IR spectra.

The results of cell studies on the novel nanofibrous materials are very important because they are directly related to the proposed applications of the material under development. From this point of view I am missing more involved discussion on the influence of material nanostructure on the cell viability. I am in doubts when reading the comment on page 60 that "The cell morphology of nanofibers with the longest hydrolytic stability (X_{DHT}Gel/$NaOC_{2:1}$ and $X_{insitu/DHT}$Gel/$NaOC_{2:1}$) were examined." because according to the figure 5.9 the most stable materials were X_{insitu}Gel/$NaOC_{2:1}$ and $X_{insitu/DHT}$Gel/$NaOC_{2:1}$.

The thesis is written in a relatively good English but there are some grammar mistakes, sometimes repeated through the whole text. The term carbodiimides is sometimes used as "carbodimides". Some figure captions do not contain all the important information.

In spite of few unclear points and the remarks on the text layout I found the submitted PhD thesis interesting and containing valuable results on electrospinning of gelatine/NaOC and PCL/gelatin filled with HNTs. This is also documented by already published paper, presentation at conferences and patent application. Therefore, I propose to accept the submitted thesis as the PhD thesis of the candidate.

Brno, Dec 7th 2016 Assoc. Prof. Lenka Zajíčková

Questions for PhD defense:

- Can you explain the physical and/or chemical principles behind the influence of humidity of the electrospinning process?
- Clarify the concentration of gelatin in acetic acid for the results shown in figure 5.2.
- Is the IR spectrum of gelatin nanofibers the same as of gelatin? What is the position of amide I and II absorption bands in pure gelatin and how is it changing with the addition of NaOC?
- Can you comment on the possible presence of Si-OH in HNTs powder as assessed by IR spectroscopy? The position of Si-OH can be around $930\,\mathrm{cm}^{-1}$.