

Opponent's Review of the Bachelor's Thesis

Isolation and characterisation of light harvesting complexes from bacteria of the genus *Gemmatimonas*

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In this thesis, the author attempts to isolate and characterize the light-harvesting complexes from the *Gemmatimonas phototrophica* AP64T which belongs to the seventh phylum of the evolutionarily important (bacterio)chlorophyll-containing bacteria. The purification of the isolated complexes was achieved by gel-filtration and electrophoresis. High-performance liquid chromatography was used for pigment identification of the isolated light-harvesting complexes. The protein subunits of these complexes were further identified by Mass spectrometry analysis.

The theoretical background was clearly summarized in introduction chapter (6 pages) which describe bacterial photosynthetic processes and also gram negative bacterium *Gemmatimonas phototrophica* AP64T. At the end of this chapter are written purposes and objectives of this work. I'd like to ask a question here.

Question 1.: One of the objective is " optimization of the cultivation process with the aim of growing a larger amount of bacteria". Did you try some "cultivation experiments" with different cultivation conditions? The text of the thesis does not mention about optimization of the cultivation process.

In "methods" chapter (7 pages) author described a general introduction to the methods and a detailed procedures witch was used in this work.

Question 2.: 2.5 Gel filtration. Description of solubilisation of membranes. What was the final concentration of n-Dodecyl β -D -maltoside? This is not clear from the text (15% stock solution, or final concentration?). Can you explain the solubilisation process and difference between n-Dodecyl β -D -maltoside and Triton?

The next chapter (6 pages) summarized results of the thesis. This chapter is nicely supplemented with figures of absorption spectra, HPLC chromatograph, CN and SDS-PAGE gels of isolated samples from studied photosynthetic bacteria. Last two

figures (15. 16.) shows models of two identified proteins of antenna-RC complex (by MS).

Question 3.: Where is figure 11? In this thesis gel filtration and a native gel electrophoresis was used. Do you know some other separation method? Which one?

The last chapter (2 pages) discuss about obtained results. The spectroscopic properties of the isolated membranes were similar to those of some other species of phototrophic bacteria. However, the position of the near-IR BChl-a Qy bands was rather unusual. To minimize the possibility that more complexes give rise to near-IR unusual bands, weaker and stronger detergents was applied for gel filtration. Also a native gel electrophoresis as a another separation method was used. With both methods two main bands were obtained, one with carotenoid-only spectrum (unknown function), and the other with the spectrum of the light-harvesting complex (supported by the MS analysis), both with near-IR bands.

The citations (3 pages) of the literature are well used in the theoretical background, as well as in the rest of the text. The cited literature is relevant and the citations are also formally correct.

This bachelor thesis shows that some photosynthetic features of *G. phototrophica* AP64T are different from those of the other known phototrophic bacteria, that makes it an interesting object to next study. In spite of the some mistakes, I **recommend** this thesis for defense with a proposed grading "1".