



Přirodovědecká
fakulta
Faculty
of Science

Jihočeská univerzita
v Českých Budějovicích
University of South Bohemia
in České Budějovice

OPPONENT'S REVIEW ON BACHELOR/DIPLOMA* THESIS

Name of the student: Linda Jernej

Thesis title: Genome analysis of *Kutzneria* sp. strain BCCO 10_1627 and detection of antifungal secondary metabolites

Supervisor: Dr.nat.techn. Erika Corretto, RNDr. Alica Chroňáková, Ph.D.

Referee: Ing. Miroslav Petříček, Ph.D.

Referee's affiliation: 1st Faculty of Medicine, Charles University, Prague

	Point scale ¹	Points
(1) FORMAL REQUIREMENTS		
Extent of the thesis (for bachelor theses min. 18 pages, for masters theses min. 25 pages), balanced length of the thesis parts (recommended length of the theoretical part is max. 1/3 of the total length), logical structure of the thesis	0-3	3
Quality of the theoretical part (review) (number and relevancy of the references, recency of the references)	0-3	3
Accuracy in citing of the references (presence of uncited sources, uniform style of the references, use of correct journal titles and abbreviations)	0-3	3
Graphic layout of the text and of the figures/tables	0-3	3
Quality of the annotation	0-3	3
Language and stylistics, complying with the valid terminology	0-3	2
Accuracy and completeness of figures/tables legends (clarity without reading the rest of the text, explanation of the symbols and labeling, indication of the units)	0-3	2
Formal requirements – points in total		19
(2) PRACTICAL REQUIREMENTS		
Clarity and fulfillment of the aims	0-3	2
Ability to understand the results, their interpretation, and clarity of the results, discussion, and conclusions	0-3	2
Discussion quality – interpretation of the results and their discussion with the literature (absence of discussion with the literature is not acceptable)	0-3	2
Logic in the course of the experimental work	0-3	2

* Choose one

¹ Mark as: 0-unsatisfactory, 1-satisfactory, 2-average, 3-excellent.

Completeness of the description of the used techniques	0-3	3
Experimental difficulty of the thesis, independence in experimental work	0-3	3
Quality of experimental data presentation	0-3	2
The use of up-to-date techniques	0-3	3
Contribution of the thesis to the knowledge in the field and possibility to publish the results (after eventual supplementary experiments)	0-3	2
Practical requirements – points in total		21
POINTS IN TOTAL (MAX/AWARDED)	48	40

Comments of the reviewer on the student and the thesis:

The presented thesis, dealing with the investigation of *Kutzneria sp. strain BCCO 10_1627* genome analysis, is divided into two major themes as there is already obvious from the title. The first one concerns the whole genome sequencing and its analysis, the second one intends to make the first preliminary study on antifungal activities produced by the respective strain.

As to the formal aspects, the work is well written and fulfills the necessary standards. I would just point out quite frequent omitting of commas, what made the reading in some parts a bit difficult.

The literature review part of the work, written on 8 pages, is well arranged and makes a nice and brief but condensed introduction to the analytical and experimental section of the work. After reading this part of thesis, it becomes apparent why the strain was chosen. The work brings the only second complete sequence of the *Kutzneria* genome after *K. albida*, while both of them belong to the genus capable of synthesis of industrially and pharmaceutically relevant secondary metabolites. I have the only one note to this section: the author should have explained to the reader less acquainted with this issue, what is the short chain length factor, how it differs from the 'normal' chain length factor and what the term chain length factor stands for.

In-silico analyses exploiting several analysis tools were thoroughly done and resulted in completely assigned genome sequence of *Kutzneria sp.* I have just question to the table 7. What the numbers in the first column mean and how many any other clusters are detected in the genome. There were mentioned 11 clusters in the table plus 57 of other BGCs. This does not correspond to numbers in the table's 1. column.

I understand that the experimental part related to antifungal activities should have brought the first preliminary results on the presence of such activities. The first part involving primary screening with living strains was well done. I have some more comments and even reproach for the second part of screening that was using extracts. In order to obtain more relevant results, it should have been requirable to apply extracts in different amounts, not only in one. Otherwise, as in this case, the amount of active compounds could have been well below the minimal inhibitory concentration in the whole zone around discs. Therefore, it is difficult to explain the difference between the activity of 'liquid' fraction sample and the absence of any activity for other fractions. This applies particularly for MeOH extractions (as e.g. table 10). Also, why in the Fig. 10 some plates are shown from the top, while some others from the bottom. Is there any purpose behind it? In some cases, e.g. concerning the strain 609, the inhibition by GYM+gluc extract can hardly be evaluated since the target colony is not grown enough and is still circular. This could have been caused by only relatively short time of cultivation and not by any interaction in contrast to the case of neighboring plate.

Since there is some discrepancy between results with liquid cultures and other fractions, there comes to the mind a question where there have gone the active compounds during the fractionation

process? It might have been that the activity in the liquid culture comes actually from the strains again growing on the agar medium together with fungi. The conclusion stated by the last sentence on the page 33 is a pure speculation, not proven by any rational experimental data (e.g. by co-cultivation of the cultures in liquid medium followed by the comparison against not co-cultivated strains). It is also questionable to use CHCl₃ as a final solvent since it is a quite non-polar and therefore not all potentially active compounds could dissolve well in it. Therefore I fully agree with the sentence on the page 32: 'The second antifungal screening needs to be repeated...'

In spite of the above listed comments, I consider the quality of presented thesis as good and sufficient to be recommended for the Bc. degree defense.

Suggestions and questions, to which the student has to answer during the defense.
Mistakes, which the students should avoid in the future:

Please see the above.

Conclusion:

In conclusion, I

r e c o m m e n d / ~~d o n o t r e c o m m e n d~~*

the thesis for the defense and I suggest the grade .²

In Praha date September 13, 2019

Miroslav Petříček

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signature



² You can suggest a grade, which can be modified during the defense based on the presentation. However, if the reviewer is not present at the defense, the grade will not be counted. Grades: excellent (1). Very good (2), Good (3), Unsatisfactory/failed (4).